DANISH CONTRIBUTION TO THE EU WATER FRAMEWORK DIRECTIVE INTERCALIBRATION PHASE 2

Technical Report from DCE – Danish Centre for Environment and Energy No. 37 2014

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Data sheet

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Abstract: This report presents the Danish contributions to the European Water Framework Directive intercalibration phase 2. The project was initiated and financed by the Danish Nature Agency and focused on development of phytoplankton indicators based on biovolume and carbon biomass, further development and adjustment of the Danish benthic fauna index, and evaluating the possibilities of intercalibrating indicators of benthic vegetation. The results of this project have been included in the Baltic GIG and NEA GIG Milestone 6 reports.

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Nota bene: Rapporten opsummerer det danske bidrag til et projekt omhandlende interkalibrering af planteplankton, bundfauna og bundvegetation mellem Danmark, Tyskland og Sverige. Analysearbejdet blev afsluttet og afrapporteret i 2011, men rapporten er først offentliggjort i maj 2014 som følge af en fejl hos DCE – Nationalt Center for Miljø og Energi.

Nota bene: The report presents the Danish contributions to a project on intercalibration of phytoplankton, benthic fauna and benthic vegetation between Denmark, Germany and Sweden. The analyses were finalized and reported in 2011, but the report itself was not published until May 2014 due to a mistake in DCE – Danish Centre for Environment and Energy.

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1 Summary

This report presents the Danish contributions to the European Water Framework Directive intercalibration phase 2. The project was initiated and financed by the Danish Nature Agency and focused on development of phytoplankton indicators based on biovolume and carbon biomass, further development and adjustment of the Danish benthic fauna index, and evaluating the possibilities of intercalibrating indicators of benthic vegetation. The results of this project have been included in the Baltic GIG and NEA GIG Milestone 6 reports.

Phytoplankton indicators were established for total phytoplankton biovolume and carbon biomass. The indicators were derived from relationships between concentrations of total nitrogen (TN) and biovolume or carbon biomass of the phytoplankton community. For each indicator site-specific ecological status class boundaries were determined.

The sub-elements 'blooms' and 'abundance' are not included in the Danish assessment systems. Blooms due to requirement of generally large data sets with high sampling frequency compared to the available data, and abundance due to the lack of indicator species which can be expected to correlate with the predominant pressure 'nutrients'.

The Danish benthic fauna index was modified to be useful at low salinities also, and methods for establishing ecological status classes and assessment of ecological status were described. For the intercalibration of type NEA8b 35 Danish stations were chosen and 11 of these were defined as benchmark stations.

Relationship between the Danish indicator 'depth limit of eelgrass' (or calculated EQRs for this indicator) and environmental parameters like TN and Secchi depht were demonstrated.

There is no joint Danish assessment tool combining depth distribution of eelgrass, cover of macrophytes, and number of perannual species. However each of those single elements has been tested against nitrogen concentration or nitrogen load which is regarded as the most important anthropogenic pressure in inner Danish waters.

It was not possible to find a common metric for intercalibration between Denmark and Sweden.

2 Sammenfatning

Denne rapport sammenfatter det danske bidrag til Vandrammedirektivets (VRD) interkalibrering fase 2 som defineret i projektet 'Udvikling og afprøvning af værktøjer til interkalibrering af planteplankton, bundfauna og makrovegetation i de danske kystområder' indgået mellem Naturstyrelsen og DCE - Nationalt Center for Miljø og Energi.

Planteplankton

Med udgangspunkt i sammenhængen mellem koncentrationen af total kvælstof (TN) og hhv. det totale biovolumen eller den totale kulstofbiomasse af planteplankton, blev der opstillet forslag til område-specifikke klassegrænser for de to planteplanktonindikatorer 'biovolumen' og 'carbon biomasse'. De definerede klassegrænser korrelerede med tilsvarende klassegrænser for klorofyl udviklet i et tidligere projekt.

De to delelementer 'blooms' og 'abundans' specificeret under det biologiske kvalitetselement planteplankton i VRD er ikke anvendt i Danmark. Der er tidligere foreslået en statistisk definition af blooms som klorofylværdier, der ligger uden for normalfordelingen i et givent område, men anvendelse af denne indikator kræver meget store datasæt med høj målefrekvens, der sjældent er til rådighed. Tilsvarende er det vurderet, at indikatoren 'abundans' ikke er anvendelig. I nogle andre lande med beskedne overvågningsdata er den foreslået anvendt på hele planteplanktonsamfundet, hvilket er meningsløst, da abundans i så fald dækker celletæthed af organismer, hvis størrelse varierer med flere størrelsesordener, og da koblingen mellem næringsrigdommen og planteplankton vil afspejles i primært biomassen eller det samlede planteplanktonvolumen. Indikatoren abundans vil kun give mening, hvis den kan anvendes på forekomsten af en enkelt art. Der er endnu ikke i danske områder fundet en sådan anvendelig indikatorart.

Bundfauna

Det danske bundfaunaindeks, DKI-indekset, blev modificeret med henblik på også at fungere ved lavere salinitet. I indekset er der nu indbygget relationer mellem salinitet og maximal Shannon-diversitet og mellem salinitet og minimumværdi af AMBI-indekset. Indekset, benævnt DKIver2, kan nu antage værdier mellem 0 og 1 i hele salinitetsintervallet 8-33 psu, og er derfor at betragte som en sand EQR.

Metoder til grænsesætning af økologisk kvalitet er beskrevet ligesom metode til vurdering af status i vandområder.

Der blev vist signifikante relationer mellem DKIver2 og presfaktoren Clostridium som indikator for spildevand, og med organisk materiale i sedimentet målt som glødetab fra Aarhus Bugt-gradienten.

I forbindelse med interkalibreringen i type NEA8b er der udvalgt 35 Danske stationer, hvoraf de 11 er udpeget som benchmark-stationer funderet på en række proxy-baserede kriterier.

Ved interkalibreringen i Baltic GIG er der udpeget tre vandområder i type BC6 og seks vandområder i type BC8 som benchmark 'sites' baseret på et snævert vindue i BSPI (Baltic Sea Pressure Index).

Bundvegetation

Interkalibreringen af bundvegetation mellem Danmark og Tyskland omfattede to tyske indeks:

- 'BALCOSIS' som benyttes langs åbne kyster (omfatter ålegræssets dybdegrænse, men har fokus på parametre for makroalger)
- 'ELBO' som benyttes i indre fjordsystemer (omfatter forskellige parametre for blomsterplanter og kransnålsalger)

og de danske indikatorer:

- Ålegræssets dybdegrænse
- Makroalgernes dækningsgrad, antal arter af flerårige makroalger og andel opportunister.

Grundlaget for interkalibreringen var tyske datasæt, hvor samtlige indeks og indikatorer kunne opgøres for et udvalg af områder. Referenceværdier for de tyske indeks var til rådighed for samtlige områder, mens referenceværdier for de danske indikatorer i de tyske områder blev vurderet ud fra referenceværdier for tilsvarende danske typeområder.

Interkalibreringen blev udført i flere trin, hvor det første var en sammenstilling af datagrundlag for interkalibrering af samtlige indeks/indikatorer. Resultatet af dette første trin var:

- Datagrundlaget var tilstrækkeligt til at gå videre med interkalibrering mellem hvert af de tyske indeks og den danske indikator 'ålegræssets dybdegrænse'.
- Derimod var der ikke tilstrækkeligt datagrundlag til at gå videre med interkalibreringen af de tyske indeks i forhold til de danske makroalgeindikatorer, idet 1) der ikke er defineret brugbare referenceværdier for de danske algeindikatorer i åbne områder, hvor BALCOSIS især bliver benyttet, og 2) der ikke var data for makroalger i de indre fjordområder, hvor ELBO især bliver benyttet.

Andet trin i interkalibreringen var korrelationsanalyser mellem EQR-værdier baseret på tyske indeks, og EQR baseret på ålegræssets dybdegrænse opgjort for de samme områder. Resultatet af dette trin var:

- EQR baseret på det tyske ELBO-indeks og den danske dybdegrænseindikator var signifikant korreleret, og det var derfor muligt at gå videre med interkalibreringen af disse.
- EQR baseret på det tyske BALCOSIS-indeks og den danske dybdegrænseindikator var ikke signifikant korreleret, og det var derfor ikke muligt at gå videre med interkalibreringen af disse.

Tredje trin i interkalibreringen var vurdering af opfølgende muligheder under 'option 2' og 'option 3', hvor:

- Option 2: Anvendes når landene ikke benytter identiske metoder/indeks, og der ikke er basis for brug af begge indeks på et fælles datamateriale. I det tilfælde skal den videre interkalibrering baseres på nationale datasæt, men i forhold til en fælles biologisk parameter.
- Option 3: Anvendes når landene ikke benytter identiske metoder/indeks, men der er basis for brug af begge indeks på det fælles datamateriale.

Da der ikke var en signifikant korrelation mellem EQR-værdierne for det danske ålegræs-indeks og BALCOSIS-indekset, var der kun mulighed for at gå videre i interkalibreringsproceduren i forhold til ELBO-indekset. Den videre interkalibrering af ålegræs-indekset og ELBO-indekset var desuden kun mulig inden for det tyske datamateriale, da flere af de parametre, som indgår i ELBO-indekset, ikke overvåges i Danmark.

- Alternativ option 2: Som udgangspunkt skal interkalibreringen baseres på en fælles biologisk parameter. Da der ikke var nogen brugbar fælles biologisk parameter anvendtes i stedet de pseudo-biologiske parametre TN og Secchi-dybde. Det danske indeks korrelerede signifikant med Secchi-dybden, mens det tyske indeks korrelerede signifikant med både TN og Secchi-dybden. Det var dog ikke muligt inden for rammerne af dette projekt at lave øvelsen inden for den samme kystvandstype (M2), hvorfor 'option 2' blev opgivet. Når denne øvelse ikke umiddelbart kunne lade sig gøre, skyldes det, at de danske data var baseret på 'vandplansperioden 2001-05', hvilket gav for få data for kystvandstypen M2, mens de tyske data var for enkelte år. Etablering af de danske data for enkelte år, ville kræve en større arbejdsindsats med at fremskaffe dybdegrænser for de enkelte år i de danske lokaliteter inden for kystvandstypen M2 samt nye udtræk af TN og Secchi-dybde og endelig genberegning på baggrund af de nye data.

- Alternativ option 3: Som udgangspunkt skal interkalibreringen ske med udgangspunkt i et fælles datamateriale. Det var som nævnt ikke muligt, men tilgangen blev alligevel forsøgt afgrænset til det tyske datamateriale. Desuden var det nødvendigt i det tyske materiale at se bort fra områder uden ålegræs, da det danske indeks er baseret på ålegræssets dybdegrænse. Med baggrund i dette var der en signifikant korrelation mellem ELBO EQR og TN men ikke mellem ELBO EQR og Secchi-dybde. Den videre analyse indikerede, at den danske EQR for overgangen mellem god og moderat skulle justeres for at matche den tyske EQR.

Konklusionen var, at det inden for rammerne af projektet ikke var muligt at lave en fuld interkalibrering mellem det danske og det tyske indeks, som fulgte den officielle procedure gennem hele analysen. I stedet blev forsøgt alternative tilgange til interkalibreringen, hvis resultat skal tages med et vist forbehold. EQR-indekset for de danske og de tyske områder tilhørte for langt størstedelens vedkommende kategorien dårlig eller moderat. De nationale korrelationer mellem EQR og TN indikerede, at for at opnå god status skal koncentrationen af TN være under 20 µmol/l.

I dag findes der ikke et dansk tilstandsvurderingsværktøj som kombinerer de eksisterende værktøjer 'dybdeudbredelse af ålegræs', 'dækning af makroalger' og 'antal flerårige arter'. Hver af disse indikatorer har vist sig at respondere signifikant på kvælstofkoncentrationer og lys.

Det danske makroalgeværktøj er i sin nuværende opsætning ikke anvendeligt til at beskrive tilstanden i åbne kystnære farvande. Værktøjet er tilpasset en beskrivelse af forholdene i fjorde og ikke åbne farvande. Projektet rummede ikke ressourcer til at tilpasse og teste værktøjets anvendelighed i åbne farvande

Undervandsvegetation i den danske type OW3 kunne ikke interkalibreres med den svenske typologi 7, da det ikke var muligt at finde et egnet datasæt.

Det danske værktøj er som anført ovenfor ikke tilpasset en beskrivelse i åbne farvande, så resultaterne for Køge Bugt og Hjelm Bugt er ikke anvendelige.

I Sverige anvendes dybdeudbredelse af udvalgte arter som biologisk kvalitetselement. Danske makroalgedata indsamles i 2 m intervaller, hvilket introducerer en stor usikkerhed i fastlæggelse af den danske dybdegrænse, men mangel på substrat er ofte den begrænsende faktor for algernes dybdeudbredelse i kystnære danske farvande. Den svenske metode er ikke testet på danske data.

Ålegræs er registreret i den svenske typologi 7, men data om dybdeudbredelse, som indgår i det danske værktøj, er ikke indsamlet i Sverige. Der er heller ikke etableret klassegrænser for ålegræs i svensk farvand.

Det var således ikke muligt at finde en fælles 'metric', der kunne anvendes til en dansk-svensk interkalibrering af bundvegetation.

3 Phytoplankton

3.1 Development of phytoplankton biovolume and carbon biomass indicators for Danish coastal waters

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.

During the initial WFD intercalibration the biological quality element phytoplankton was intercalibrated using the concentration of chlorophyll a as an indicator of phytoplankton biomass only. According to the WFD assessments using phytoplankton should include, in addition, metrics relating to phytoplankton taxonomic composition, abundance, and blooms.

Danish waters are located in the transition zone between the brackish Baltic Sea and the saline North Sea. The salinity gradient across the Danish monitoring stations and salinity gradients in general is a main factor influencing the taxonomical composition of the phytoplankton (Gasiunaite et al. 2005, Henriksen et al. 2011). Thus, the optimal development of indicators would be site-specific within a narrow salinity range. However, only a very limited number of monitoring stations with data on taxonomical composition of phytoplankton are available and the within-station gradients of pressures (nutrient loads, nutrient concentrations) are generally insufficient to establish site-specific dose-response relationships. Therefore the development of indicators and establishing of reference conditions via dose-response relationships has been based on combined data sets covering all available stations. This has led to suggested indicators based on the total biomass of phytoplankton while attempts to include taxonomical composition of the phytoplankton proved unsuccessful.

In the following result of development of Danish phytoplankton indicators is presented.

3.1.1 Data sets

Data included in this project originate from the Danish national monitoring programme. Data have been compiled from the present database for environmental data from surface waters (ODA) combined with data from the former national database for marine data (MADS) hosted at Aarhus University. Data sets from the period 1988 to 2009 were included in the analyses. The stations were selected based upon sampling frequency and so that the stations provide a wide gradient in the environmental parameters studied (Carstensen et al., 2008).

Phytoplankton samples from the monitoring program were integrated samples (surface to 10 m depth or surface down to 0.5 above the sea bed in shallow water stations) preserved in Lugols' solution and enumerated in the inverted microscope according to HELCOM procedures (HELCOM 1988). Organisms were identified to species, genus or class/group (in case of specimens too small to identify in the microscope) and the dimensions of approx. 10 specimens of each counted taxon were measured for calculation of biovolume. Phytoplankton carbon biomass was calculated from cell counts and dimension measurements assuming simple geometric shapes and using conversion factors of 0.13 and 0.11 pg C µm-3 for thecate dinoflagellates and other phytoplankton groups, respectively. Carbon contents of diatoms were corrected for lower C content of cell vacuoles [pg C (µm3 vacuole)-1 = 0.1 * pg C (µm3 plasma volume)-1] according to Edler (1979).

Class boundaries for the indicator 'chlorophyll' originate from Carstensen et al. (2008).

3.1.2 Statistical analyses

Yearly values of plankton biovolume and nutrient concentrations were estimated by applying a generalised linear model to data from each station separately:

$$E(y) = b_0 + b_1 month + b_2 year + e$$

The LSmeans were estimated for years and used in the subsequent analysis.

Biovolumes of plankton were estimated as summer means of the auto- and mixotrophic plankton community based upon samples taken between May and September.

For each year values for concentrations of total N (TN) were estimated based upon measurements taken within the first six month (January-June) of the year.

Reference conditions and boundary values for biovolume and carbon biomass were calculated from the biovolume-TN or carbon biomass-TN regression using the corresponding values for TN as input. TN values were obtained from Carstensen et al. (2008). Standard errors for these estimates were found by Monte Carlo simulation taking variations in the estimated model as well as uncertainty of the TN reference condition and boundary values into account.

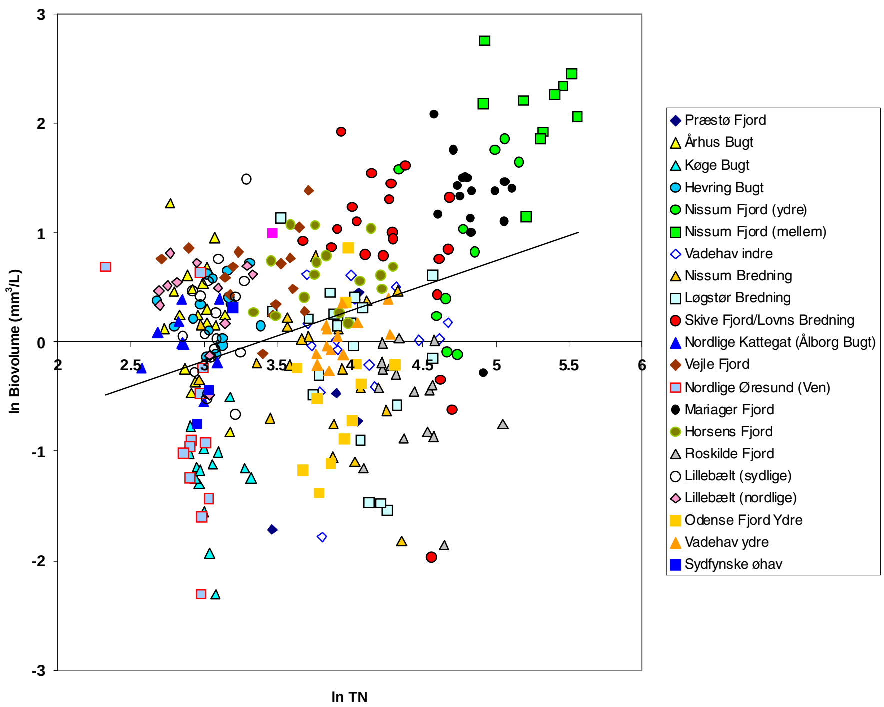
3.1.3 Results

Results of the data analyses revealed significant relationships between the eutrophication indicator TN and the total biovolume or carbon biomass of phytoplankton.

Biovolume

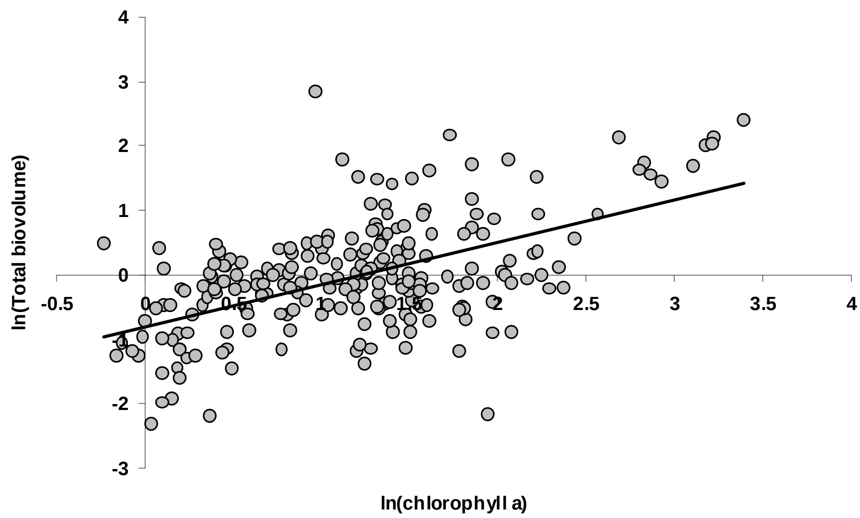
A significant relationship between TN and total summer biovolume of the phytoplankton was found (Fig. 3.1).

Figure 3.1. Total summer biovolume of plankton community versus winter-spring TN mean concentrations for 21 different sites.



In addition, the total biovolume of phytoplankton was significantly correlated with the summer mean concentration of chlorophyll a. This relationship was, however, characterised by large scatter (Fig. 3.2).

Figure 3.2. Total summer biovolume of phytoplankton community versus summer mean chlorophyll a concentrations for the 21 different sites presented in Fig. 3.1.



Based on the TN-biovolume relationship and a combination of hind-casted estimates of N-inputs to the inner Danish waters and expert judgement of the corresponding ecological status during different time periods, WFD compliant class boundaries were established for biovolume at these stations (Table 3.1). Reference conditions and boundary values were calculated from the TN-biovolume regression using the corresponding reference and boundary values of TN as input (Carstensen et al. 2008). Standard errors for these estimates were found by Monte Carlo simulation taking variations in the estimated model as well as uncertainty of the TN reference condition and boundary values into account.

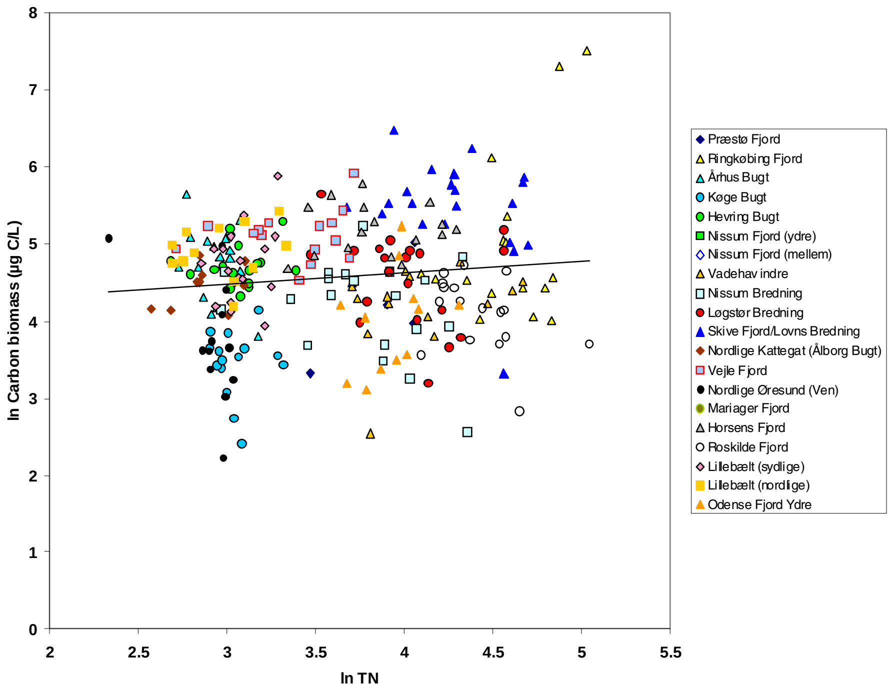
Table 3.1. Suggested reference conditions and boundary values for summer (May-September) phytoplankton biovolume (mm^3/L) computed from corresponding values of TN concentrations (Carstensen et al. 2008). Boundary values between good and moderate status are highlighted. Stations marked with * are located within the Baltic GIG. All other stations are located within the NEA GIG.

	Ref. cond.	H/G	G/M	M/P	P/B
Hevring Bugt	0.69 (±0.05)	0.73 (±0.06)	0.81 (±0.08)	0.89 (±0.11)	0.94 (±0.13)
Horsens Fjord	0.80 (±0.05)	0.93 (±0.06)	1.16 (±0.07)	1.41 (±0.10)	1.55 (±0.11)
Køge Bugt *	0.69 (±0.05)	0.73 (±0.05)	0.79 (±0.07)	0.87 (±0.08)	0.92 (±0.09)
Lillebælt *	0.69 (±0.05)	0.72 (±0.05)	0.79 (±0.06)	0.86 (±0.07)	0.91 (±0.07)
Løgstør Bredning	0.91 (±0.05)	1.12 (±0.06)	1.49 (±0.09)	1.86 (±0.13)	2.08 (±0.16)
Nissum Bredning	0.83 (±0.05)	0.98 (±0.06)	1.24 (±0.08)	1.51 (±0.10)	1.67 (±0.13)
Nissum Fjord	1.25 (±0.06)	1.69 (±0.10)	2.42 (±0.19)	3.14 (±0.32)	3.56 (±0.39)
Nordlige Kattegat	0.69 (±0.05)	0.72 (±0.06)	0.78 (±0.07)	0.85 (±0.09)	0.90 (±0.11)
Odense Fjord Ydre	0.92 (±0.05)	1.14 (±0.06)	1.51 (±0.09)	1.89 (±0.13)	2.12 (±0.16)
Præstø Fjord *	0.86 (±0.06)	1.03 (±0.08)	1.33 (±0.12)	1.64 (±0.17)	1.82 (±0.20)
Roskilde Fjord	0.90 (±0.05)	1.11 (±0.06)	1.46 (±0.08)	1.83 (±0.12)	2.04 (±0.14)
Skive Fjord/Lovns Bredning	0.98 (±0.05)	1.23 (±0.06)	1.67 (±0.09)	2.12 (±0.15)	2.38 (±0.18)
Sydfynske Øhav *	0.71 (±0.05)	0.76 (±0.06)	0.87 (±0.07)	0.97 (±0.08)	1.04 (±0.09)
Vadehav indre	0.86 (±0.05)	1.04 (±0.06)	1.35 (±0.07)	1.67 (±0.10)	1.85 (±0.13)
Vadehav ydre	0.82 (±0.06)	1.14 (±0.06)	1.66 (±0.09)	2.17 (±0.15)	2.46 (±0.20)
Vejle Fjord	0.75 (±0.05)	0.84 (±0.06)	1.00 (±0.07)	1.17 (±0.09)	1.27 (±0.10)
Århus Bugt	0.67 (±0.05)	0.70 (±0.05)	0.74 (±0.06)	0.79 (±0.07)	0.81 (±0.08)
Øresund Nord	0.68 (±0.05)	0.71 (±0.05)	0.76 (±0.05)	0.82 (±0.06)	0.86 (±0.06)

Carbon biomass

The relationship between TN and phytoplankton carbon biomass is shown in Fig. 3.3.

Figure 3.3. Carbon biomass of phytoplankton community versus winter-spring TN mean concentrations.



Based on the TN-carbon biomass relationship and a combination of hindcasted estimates of N-inputs to the inner Danish waters and expert judgement of the corresponding ecological status during different time periods, WFD compliant class boundaries were established for carbon biomass at these stations (Table 3.2).

Table 3.2. Suggested reference conditions and boundary values for summer (May-September) phytoplankton carbon biomass (µg C/L) computed from corresponding values of TN concentrations (Carstensen et al. 2008). Boundary values between good and moderate status are highlighted. Stations marked with * are located within the Baltic GIG. All other stations are located within the NEA GIG.

	Ref. cond.	H/G	G/M	M/P	P/B
Hevring Bugt	72.4 (±5.21)	74.6 (±5.30)	78.9 (±6.00)	83.2 (±6.95)	85.8 (±7.69)
Horsens Fjord	78.7 (±4.68)	85.7 (±4.64)	97.3 (±5.25)	108 (±6.86)	115 (±8.05)
Køge Bugt *	72.1 (±5.14)	74.2 (±5.09)	78.3 (±5.31)	82.3 (±5.74)	84.8 (±6.14)
Lillebælt *	72.1 (±5.13)	74.0 (±5.03)	77.8 (±5.02)	81.8 (±5.10)	84.2 (±5.29)
Løgstør Bredning	84.5 (±4.49)	95.3 (±4.83)	112 (±7.07)	127 (±10.1)	136 (±12.2)
Nissum Bredning	80.0 (±4.71)	88.0 (±4.61)	101 (±5.63)	113 (±7.44)	120 (±8.98)
Nordlige Kattegat	71.9 (±5.18)	74.0 (±5.26)	77.6 (±5.60)	81.3 (±6.36)	83.6 (±6.87)
Odense Fjord Ydre	84.9 (±4.45)	95.9 (±4.79)	113 (±7.16)	129 (±10.5)	137 (±12.5)
Præstø Fjord *	81.6 (±4.89)	90.6 (±5.40)	105 (±7.25)	119 (±10.0)	126 (±11.6)
Roskilde Fjord	84.1 (±4.47)	94.6 (±4.64)	111 (±6.58)	126 (±9.79)	134 (±11.7)
Skive Fjord/Lovns Bredning	88.0 (±4.33)	101 (±5.10)	120 (±8.34)	137 (±12.4)	147 (±14.9)
Vejle Fjord	75.7 (±4.95)	80.6 (±4.74)	89.3 (±5.07)	97.7 (±5.81)	102 (±6.44)
Øresund Nord	71.5 (±5.12)	73.2 (±5.03)	76.4 (±4.93)	79.9 (±4.87)	81.8 (±4.82)
Århus Bugt	71.1 (±5.18)	72.5 (±5.15)	75.0 (±5.28)	77.7 (±5.59)	79.2 (±5.81)

Comparison of class boundaries

Danish site-specific class boundaries for chlorophyll concentrations were suggested by Carstensen et al. (2008). For evaluation of correspondence between these class boundaries and those established for phytoplankton biovolume Fig. 3.4 and 3.5 show the chlorophyll-biovolume and chlorophyll-carbon biomass class boundary relationships. There was a very strong correlation between class boundaries established for chlorophyll, biovolume and carbon biomass, respectively.

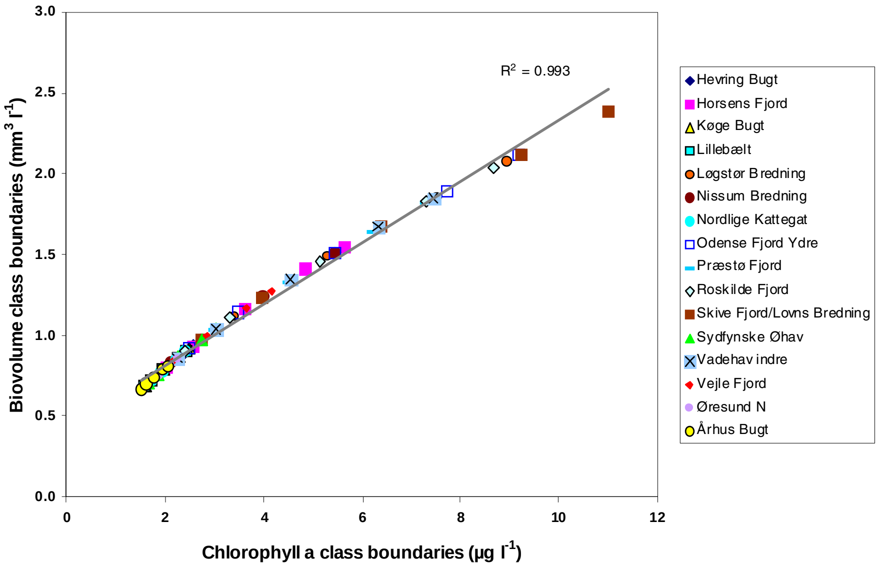


Figure 3.4. Relationship between site-specific class boundaries for chlorophyll (Carstensen et al. 2008) and corresponding boundaries for phytoplankton biovolume. For each site the reference condition and four class boundaries (tables 3.1 and 3.2) are plotted.

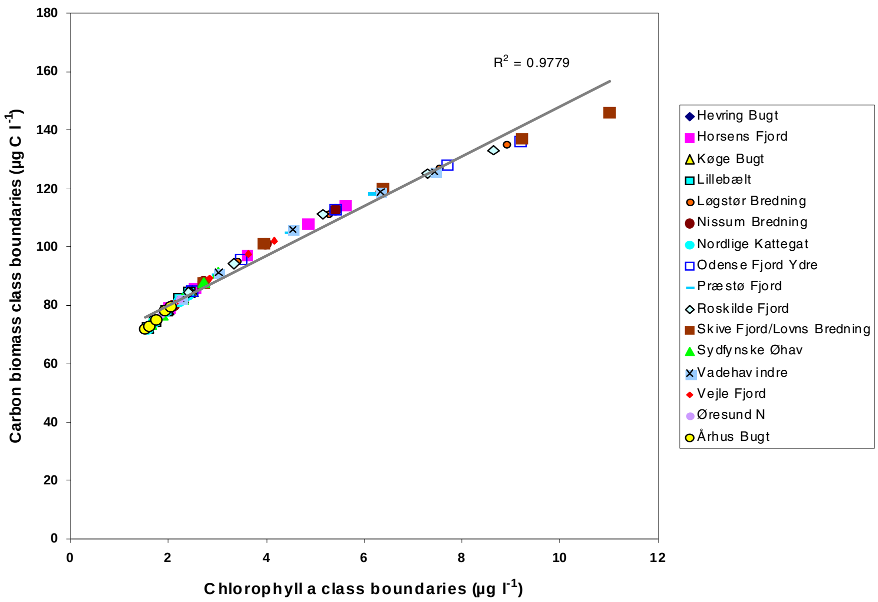


Figure 3.5. Relationship between site-specific class boundaries for chlorophyll (Carstensen et al. 2008) and corresponding boundaries for phytoplankton carbon biomass. For each site the reference condition and four class boundaries (tables 3.1 and 3.2) are plotted.

3.2 Phytoplankton blooms

Carstensen et al. (2006) proposed a definition for identification of blooms and used this definition to investigate the underlying mechanisms of summer blooms and their link to nutrient enrichment in Danish estuaries. Blooms were defined as chlorophyll a observations deviating significantly from a normal seasonal cycle; the frequency and magnitude of these deviating observations characterized bloom frequency and intensity. The definition was applied to a large monitoring data set from five estuaries in Denmark with at least biweekly sampling. Four mechanisms with links to nutrient enrichment were identified as sources of summer blooms: (1) advection from biomass-rich inner estuary, (2) resuspension of nutrients and algae from sediments, (3) nutrient releases from sediments during hypoxic conditions, and (4) decoupling of benthic grazers. Bloom frequency and intensity decreased from 1989 to 2004, corresponding to decreases in nutrient inputs and concentrations, but only bloom frequency could be directly linked to the actual total nitrogen concentrations, whereas bloom intensities depended on site-specific features, particularly a threshold response for stations exposed to hypoxia. Bloom frequency has increased over longer timescales in response to nutrient enrichment.

The identified relation with TN suggested that bloom frequency may be used as an ecological indicator in relation to eutrophication, but the complexity of bloom mechanisms, evident by the large variation around the regression line, questions if bloom frequency is also a precise indicator useful for assessing ecological status. Inclusion of site-specific features combined with data on driving forces, typically wind, may reduce the random variation, at the cost of indicator generality. Thus, the frequency of summer blooms in Danish estuaries is most likely higher today than under pristine conditions, but it will require large amounts of data and large changes in nutrient conditions to document significant changes.

The high-frequent data required for defining blooms are generally not available in the monitoring programmes of most countries and therefore this tool has been considered inapplicable to the data Baltic Sea GIG data set.

3.3 Abundance of phytoplankton

Abundance of phytoplankton has not been included in the Danish assessment system for several reasons:

1. a lack of reference sites hinders the definition of reference conditions

2. due to the huge size span in the phytoplankton species enumerated and that total volume (or the carbon biomass) rather than cell numbers relate to nutrients, the Danish focus has been on development of an assessment system based on total biovolume and carbon biomass of phytoplankton

3. the abundance/biovolume/biomass of no single species (where the problem of size differences can be almost eliminated) proved useful as an indicator of eutrophication (see also Carstensen & Heiskanen, 2007).

4 Benthic fauna

4.1 Adjusting the DKI index to attain values between 0 and 1 in all salinity regimes in the range 8-33 psu

The Danish DKI index was corrected for salinity into a new version of the index called DKI ver.2 to attain values between 0 and 1 for all salinities between 8 and 33 psu.

The "DKI "index of benthic quality was developed for use in poly- to euhaline benthic environments characterised by a relatively high species diversity (Borja et al. 2007) and has been used with success in such environments in the Northeast Atlantic area (Borja et al. 2007, Josefson et al. 2009). However when applying DKI on data from low saline and species-poor estuarine areas like the Baltic Sea area, it soon became clear that the range of possible index values was markedly restricted to the lower end of the range in saline areas. This was likely a result of salinity influence on three of the components in the index, the Shannon-wiener (H) and the number of species (S) components, but also, as we shall see, the AMBI component. It is well known that diversity of species with marine affinity decreases with decreased salinity at several spatial scales, when going from Skagerrak/Kattegat through the Belt Seas into the Baltic and further north and east (e.g. Remane 1934, Bonsdorff and Pearson 1999, Josefson and Hansen 2004, Villnas and Norkko 2011). The effect of salinity on sensitivity classification such as AMBI, however, has yet to be demonstrated.

In a comprehensive analysis of DKI in Danish waters including species data from 2600 samples of Van Veen size (0.1 m^2) from 540 sampling points (sites) there were demonstrated clear salinity effects on H, S and AMBI in open sea areas but not in closed fjords and lagoons (Josefson 2008). Maximum values of H and S decreased, and minimum values of AMBI increased, with decreasing salinity in the salinity range 8 – 28 psu.

In order to resolve the above mentioned problems with DKI in low saline areas, components in DKI is corrected for salinity as follows:

1) *The S factor (1-1/S) which becomes effective at species numbers < 10 has been omitted*. This because species numbers per 0.1 m^2 in the Baltic often is below this value also in undisturbed areas.

2) *Hmax in the Shannon-wiener factor is determined from a regression between Hmax and bottom water salinity* (Table 1).

3) *The minimum value of AMBI is determined from a regression between AMBImin and salinity and subtracted from AMBI in the original formula. If negative, AMBImin is set to 0.*

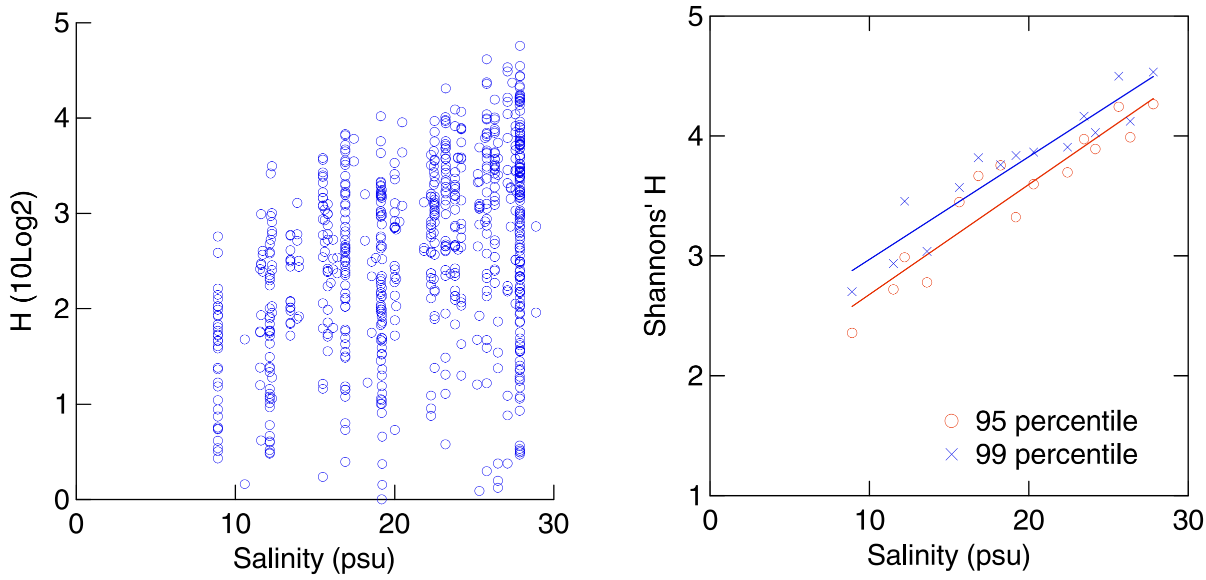


Figure 4.1. Plots of H against salinity (upper) and Hmax assessed by 99 or 95th percentiles against salinity (lower). Data from Van Veen- sized (0.1 m2) samples from meso- and poly-haline Danish open sea areas (Josefson 2008).

The regression was obtained by regressing the 99th percentile of H values from 15, approximately similar sized classes, against salinity (psu) in the interval 8-28 psu (Fig. 4.1, Table 4.1). Extrapolation up to eu-saline conditions (ca. 33 psu) gave a Hmax close to 5 which is a very reasonable max value. It was also used in the NEA GIG first round (Borja et al. 2007). Extrapolation to salinities less than 8 psu, however, is probably not relevant.

Table 4.1. Regressions between Hmax (H99) and salinity, and between AMBImin (AMBI01) and salinity.

Relation	n	Rsq	P
H99=2.117 + 0.086 * Sal (psu) (eq. 1)	15	0.89	0.000
AMBI01=3.083 – 0.111 * Sal (psu) (eq. 2)	15	0.57	0.001

The minimum AMBI (AMBImin) decreases with increasing salinity as shown in Fig. 4.2 and the reason behind is likely changes in the proportions of different sensitivity groups as shown in Fig. 4.2. At low salinities AMBI is determined to a great extent by group III (which includes *Macoma balthica*) whereas at high salinities several groups contribute to the index and group I, the "sensitive species", has the highest proportion of the individuals. AMBImin was assessed by the 1st percentile and regressed against salinity using the same salinity intervals as for H above (Fig. 4.2, Table 4.1).

The resulting formula for DKIver2 now reads:

$$\mathbf{DKI = ((1- ((AMBI-AMBImin)/7))+ (H/Hmax))/2 * (1-(1/N))}$$

where

Hmax = f (salinity), Table 4.1 eq. 1

AMBImin = f (salinity), Table 4.1 eq. 2

N = Number of individuals (as before)

The DKI is applied on 0.1 m^2 samples and therefore smaller samples like Haps has to be pooled. Results may be regarded as EQR values where the "reference" is the best value we can get at a given salinity.

By adjusting the index to salinity it can now attain values between 0 and 1 at all salinity levels in the salinity range 8 – 33 psu.

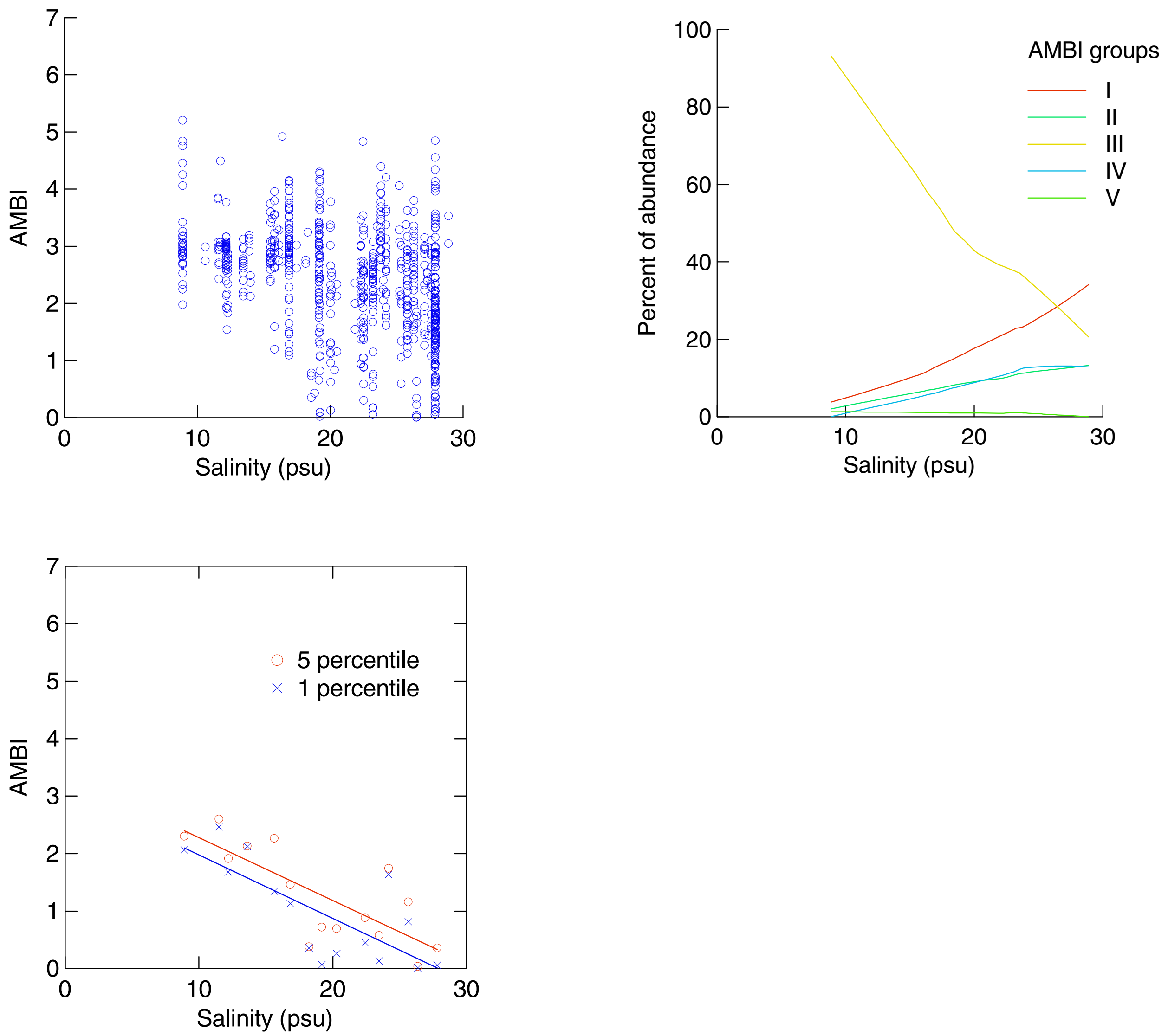


Figure 4.2. Plots of AMBI against salinity (upper graph) and AMBImin assessed by 1st or 5th percentiles against salinity (lower graph). Middle graph shows changes with salinity in proportions of the five AMBI groups of sensitivity (LOWESS lines, data points omitted for clarity). Data from Van Veen- sized (0.1 m^2) samples from meso- and poly-haline Danish open sea areas (Josefson 2008).

4.1.1 Boundary setting

Usually, the border between good and moderate EcoQS (G/M) is determined as some deviation from a reference situation. Reference data, however, are difficult to find. The Good-Moderate border for DKI was set by using the discontinuity in the relationship of anthropogenic pressure and the biological response as described in Josefson et al. (2009). The threshold value, where faunal structure deterioration commences, was identified from non-linear regression between DKIver2 and the impact proxy: distance from point source in the Aarhus Bight pollution gradient. Using a bootstrap procedure as described in Leonardsson et al. (2009) and Josefson et al. (2009) the 5th percentile of the index values from the less impacted side of the threshold was determined. It was assumed that these values represented at least Good EcoQS. The 5th percentile of these data was defined as the G/M border and attained the value of 0.68. By dividing the ranges 0-0.68 and 0.68-1 with 3 and 2 respectively the following boundary was obtained.

Poor-Bad	Moderate-Poor	Good-Moderate	High-Good
0.23	0.45	0.68	0.84

4.1.2 Water body assessment

Status in a water body is assessed by comparing the 20th percentile, which corresponds to the lower border of an 80% confidence interval, (obtained by bootstrapping, Leonardsson et al. 2009) of the DKI values calculated on individual samples from a water body with WFD boundaries set from gradient data (above). For example, for the status to be at least Good, the 20th percentile has to be above the Good-Moderate border, and then the EcoQS of the water body is acceptable.

4.2 Pressure-impact relations

Due to lack of suitable gradient data from the SW Baltic area, pressure-impact relations with DKIver2 are only described from the NEA8 area. The relations come from the sewage/urban effluent gradient in the Aarhus Bight/Samsø area.

Bacterial numbers in the surface sediments of *Clostridium* was used as an indicator of sewage impact and has been shown to change inversely to DKI with distance from the treatment plant (Josefson et al. 2009).

Similar to DKIver2 and *Clostridium*, the ignition loss of the sediment changed with distance from the treatment plant. Ignition loss is a proxy of organic matter and represents very approximately 2 times the carbon percentage.

Figure 4.3. DKIver2 showed a clear saturation curve response with increasing geographical distance from the Marselisborg sewage treatment plant / harbour of Aarhus. There were low index values close to the plant, some even in Bad status, followed by an increase up to a threshold distance of ca. 11 km and after that a levelling off.

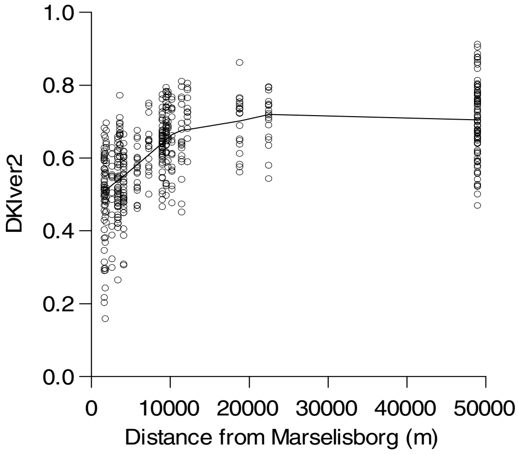


Figure 4.4. There was a highly significant nearly linear relation between DKIver2 and Log10 *Clostridium* numbers (n= 398, r= 0.314, P<0.0001).

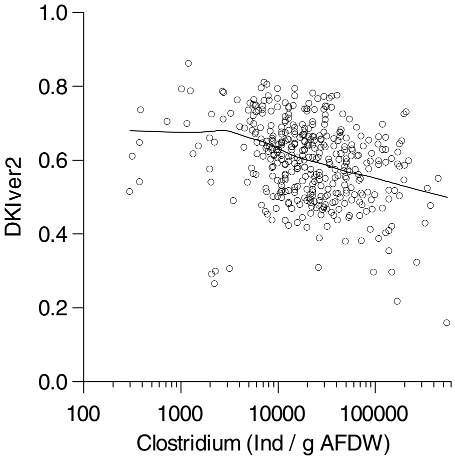
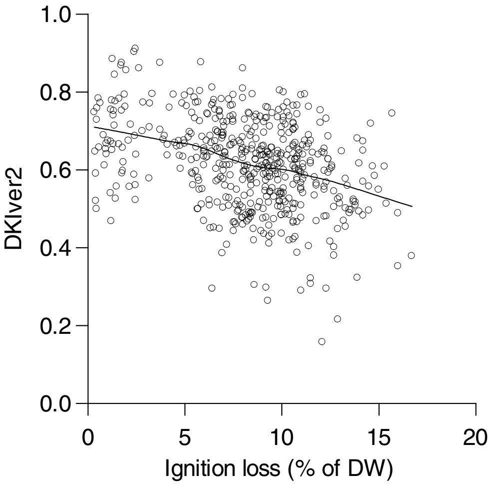


Fig. 4.5.There is a highly significant nearly linear negative relation between DKIver2 and ignition loss (n=509, r=0.374, P<0.0001).



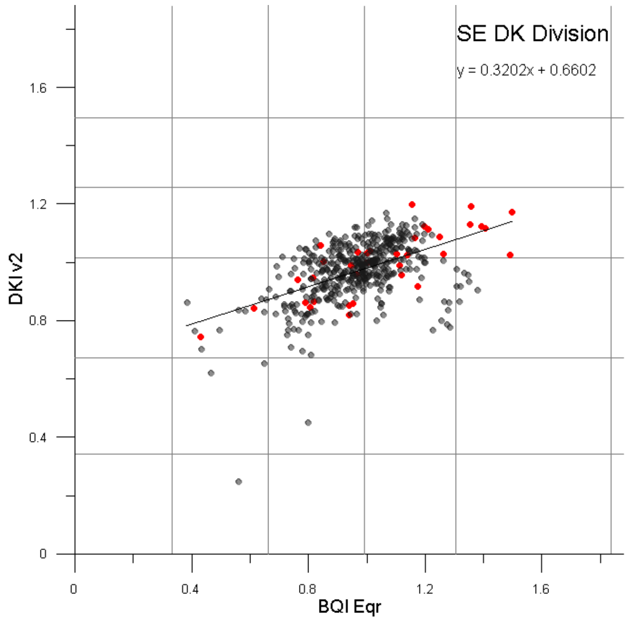
4.3 Selection of Danish data for a common data set with Norway and Sweden for NEA 8

Selection of Danish data for the second inter-calibration round of NEA8b, in all 35 stations each with three replicate Van Veen samples (0.1 m^2), were included in the inter-calibration. All stations are from water depths greater than ca. 15 m i.e. the average position of the halocline separating the saltier Kattegat deep water from the overlaying fresher Baltic water. The stations are fairly evenly distributed from the Samsø Belt area to the area north of Læsø and all were sampled in the same year, 2004.

Selection of benchmark stations

Of the Danish stations in NEA8b, 11 were chosen as benchmark sites based on the following criteria:

1. Not situated in the vicinity of pollution point sources (harbours, factories, cites etc).
2. Well ventilated areas i.e. not situated in basins, but rather on the even slope towards the open sea.
3. No reports of oxygen deficiency during the previous 10 years
4. Situated within a distance-interval of 1-10 km (NO 0-10 km) from the coast.
5. Situated in the depth interval 25-50 m (NO >25m).



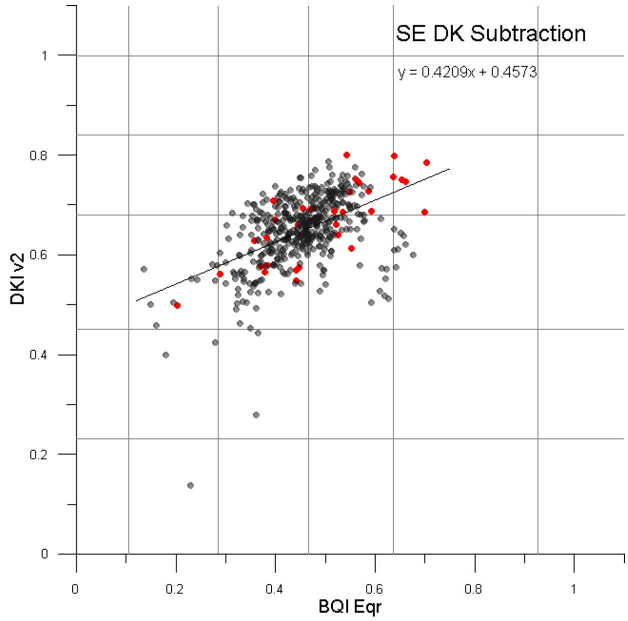


Figure 4.6. Relationships between Swedish and Danish indices after benchmarking were significant using both subtraction and division methods (red dots=Danish sites, and black dots=Swedish sites).

Member State/Method	r	p
SE BQI vs. DK DKIver2 subtraction	0.50	<0.0001
SE BQI vs. DK DKIver2 division	0.51	<0.0001

In the Baltic GIG in typologies BC6 (SE-DK) and BC8 (DE-DK) intercalibration of status was made of water bodies each containing a number of stations. Benchmark water bodies, in the following called sites, from the pairs of countries were selected from a narrow pressure window of the Baltic Sea Pressure Index (BSPI).

BC6 (SE-DK): A BSPI range of 58.7–64.1 was used as benchmark criteria. However, no difference was found in comparability with or without benchmarking and hence benchmarking was skipped in the further analyses. The Danish benchmark sites were:

- Falster
- Fakse Bight
- Køge Bight.

BC8 (DE-DK): A BSPI range of 57–63 was used for the choice of benchmark sites. Another window of 55–65 was also tested, but did not change the results, thus was not used for further analysis.

The following water bodies were used as benchmark sites in DK:

- Rødsand (1 benchmark value)
- Flensborg Fjord, inner part + Flensborg Fjord, outer part (1 benchmark value)
- Southern Little Belt Als-Ærø (1 benchmark value)
- The Archipelago of southern Funen, open part (1 benchmark value)
- Langelandssund + Great Belt 12sm, open part + Great Belt 12sm south, open part (1 benchmark value)
- Great Belt, open part + Smålandsfarvandet, open part + Smålandsfarvandet, south + Guldborgsund + Grønsund + Avnø Fjord (1 benchmark value).

5 Benthic vegetation

5.1 Compliance of Danish BQE's and validation against pressures - correlations between eelgrass EQR and TN and Secchi depth

The depth limit of eelgrass, used as indicator of ecological status in Denmark, is defined as the maximum depth of 10% eelgrass cover. The eelgrass depth limit EQRs for Danish water bodies for the period 2001-2005 (defined as: eelgrass depth limit (2001-05)/reference eelgrass depth limit), were obtained from the Nature Agency and were correlated with TN and Secchi depth levels of the water bodies (data used in correlations are presented in Table 5.1). Data on concentrations of total nitrogen (TN) and Secchi depth from 2001-2005 for Danish marine water bodies were obtained from the national marine database MADS.

Prior to analysis the data was evaluated to discard data of low value. EQRs not based on actual observations of eelgrass depth limit (2001-05) were discarded - this relates to situations where the eelgrass depth limit was modelled from TN concentrations, in some shallow areas leading to eelgrass depth limits being higher that the maximum depth of the water body. Data were also discarded if the main distribution depth of eelgrass (depth of 10% cover) from 2001-05 was more than double the Secchi depth - as this indicates that Secchi depth was obtained in an area of the water body being significantly different from the area of the eelgrass transects. Further, data were discarded if more than 25% of the Secchi depths measured in given water body equalled bottom depth - as the mean Secchi depth in these cases was significantly limited by water depth at the station and therefore not just regulated by water clarity. Finally, TN and Secchi data were discarded if there were less than four measurements per year (< 20, 2001-05) to reduce the risk of seasonal biased means - as less intensive monitoring tends to focus on the summer months. The eelgrass EQR-values obtained from the Nature Agency are mean values for each water body calculated based on a number of sites in each water body. Possible differences in handling of sites without eelgrass would affect the EQR-values of the various water bodies, but were not taken into account here as only mean values were provided.

Sorting the data as described above resulted in data sets from 46 water bodies. In these water bodies eelgrass depth limit EQRs were correlated with annual means and seasonal means (March-October, both months inclusive) of TN and Secchi depth from 2001-05. Further, mean eelgrass depth limit for 2001-05 was correlated with mean TN and Secchi depth for the same period.

Table 5.1. Danish sites included in the correlation between eelgrass EQR or eelgrass depth limit and concentration of total nitrogen (TN) or Secchi depths. EQR is the relation between present day (average 2001-2005) and reference condition of eelgrass depth limit; TN and Secchi are present day data (average 2001-2005).

Sites	**Eelgrass**		**TN (µmol/l)**		**Secchi (m)**	
	EQR	**Depth limit (m)**	**year**	**Mar-Oct**	**year**	**Mar-Oct**
Roskilde Fjord, outer part	0.61	3.4	36	32	4.7	4.6
Roskilde Fjord, inner part	0.58	2.3	66	60	4.2	4.2
Øresund, northern part	0.55	6.0	20	19	10.5	10.7
Isefjord	0.59	4.3	31	28	6.0	5.8
Sejerøbugt	0.43	5.2	20	19	7.4	7.5
Kalundborg Fjord	0.40	3.9	21	21	6.5	6.6
Smålandsfarvandet, open part	0.59	6.4	20	19	7.2	7.2
Guldborgsund	0.56	3.0	21	21	6.9	6.9
Østersøen	0.60	5.6	18	17	7.7	7.7
Gamborg Fjord	0.44	3.3	23	22	6.3	6.0
Kerteminde Fjord	0.06	0.4	28	26	5.1	4.8
Nyborg Fjord	0.33	2.8			6.0	6.0
Helnæs Bugt	0.68	5.1	24	23	6.8	6.5
Lunkebugten	0.54	3.7	20	19	5.7	5.2
Langelandssund	0.65	7.1	19	18	6.8	6.8
Det Sydfynske Øhav, open part	0.48	5.3	32	29	5.9	5.8
Odense Fjord, outer part	0.46	2.6	38	31	3.5	3.5
Lillebælt, southern part	0.39	4.2	19	18	7.6	7.6
Genner Bugt	0.24	2.8			6.4	6.3
Åbenrå Fjord	0.37	4.5	23	21	6.5	6.1
Als Fjord	0.38	4.6			6.4	5.9
Augustenborg Fjord	0.24	1.3	28	25	5.6	5.2
Haderslev Fjord	0.43	2.5			4.1	4.0
Nybøl Nor	0.36	3.3	38	32	5.0	4.6
Sydlige Lillebælt, north of Als	0.37	4.0	19	18	7.2	7.0
Flensborg Fjord, inner part	0.11	1.1	33	29	5.6	4.7
Flensborg Fjord, outer part	0.36	4.7	22	21	6.9	6.7
Sydlige Lillebælt Als-Ærø	0.49	5.3			7.9	7.9
Vejle Fjord, outer part	0.23	2.6			5.9	5.9
Vejle Fjord, inner part	0.27	2.3	23	22	4.5	4.4
Kolding Fjord, inner part	0.30	1.6	34	34	2.6	2.6
Endelave and CW [1)]outside Norsminde Fjord	0.37	4.5	18	17	6.9	6.8
Horsens Fjord, inner part	0.07	0.6	34	29	3.8	3.8
Ringkøbing Fjord	0.30	0.9	73	60	1.6	1.7
Randers Fjord, outer part	0.26	1.5	64	52	3.2	3.3
Hevring Bugt	0.27	3.3	21	18	6.7	7.0
Ebeltoft Vig	0.57	5.6			8.5	8.5
Århus Bugt syd, Samsø and Djursland Syd	0.42	5.1	18	17	8.9	8.9
Knebel Vig	0.50	4.7			7.7	8.1
Kalø Vig, inner part	0.57	5.3			8.5	8.8
Århus Bugt, Kalø and Begtrup Vig	0.52	4.9	19	17	8.4	8.5
Kattegat	0.30	3.8	17	16	7.9	8.1
Limfjorden I[2)]	0.40	2.2	43	37	3.3	3.3
Limfjorden II [3)]	0.33	1.8	56	51	3.0	2.8
Mariager Fjord, inner part	0.14	0.6	86	80	3.3	3.1
Lillebælt, northern part	0.18	2.2	19	19	6.5	6.4

[1)] CW: coastalwaters, [2)] Limfjorden I: Nissum Bredning, Thisted Bredning, Kås Bredning, Løgstør Bredning, Nibe Bredning, and Langerak, [3)] Limfjorden II: Lovns Bredning, Skive Fjord, Riisgårde Bredning, and Bjørnholms Bugt.

Eelgrass EQR correlated with TN

The Danish eelgrass depth limit EQR demonstrated only a very weak and non-significant correlation with annual and seasonal means of TN (Fig 5.1A & B, r2=0.1, p=0.1), but as expected there was a tendency towards higher EQR in water bodies with low TN. The EQR covered only the classes from bad to moderate, and due to the weak correlation it was not possible to identify levels of TN corresponding to the good/moderate boundary with acceptable certainty. Still, the data indicate that the TN concentration needs to be below 20 µmol l^{-1} to obtain a good status in most Danish water bodies.

Eelgrass EQR correlated with Secchi depth

As for TN, the Danish eelgrass depth limit EQR demonstrated a weak but significant correlation with annual and seasonal means of Secchi depth (Figs 5.2A & B, r2=0.2, p<0.01), but as expected there was a tendency towards higher EQR in water bodies with high Secchi depth. The EQR covered only the classes from bad to moderate, and due to the weak correlation it was not possible to identify levels of Secchi depth corresponding to the good/moderate boundary with acceptable certainty. Still, the data indicate that the Secchi depth in most water bodies needs to be more than 7 m to obtain a good status in most Danish water bodies.

Eelgrass depth limit as a function of TN

The eelgrass depth limit from 2001-05 correlated highly significantly with concentrations of TN from the same period (Figs 5.3A & B, r2<0.0001, p=0.4). The eelgrass depth increased with decreasing TN. However, there was a high variation of eelgrass depth limits at low TN. Further, there was a very steep decrease in eelgrass depth limit with increasing TN up to 30-40 µmol l^{-1} as opposed to a very weak response in eelgrass depth limits at higher TN levels. TN levels of 30-40 µmol l^{-1} thus seem to be a kind of N-response threshold across Danish water bodies at least for the period 2001-2005. Therefore, the relation between eelgrass depth limit and TN would be better described by a non-linear function or by a linear function on each side of the 'threshold' value.

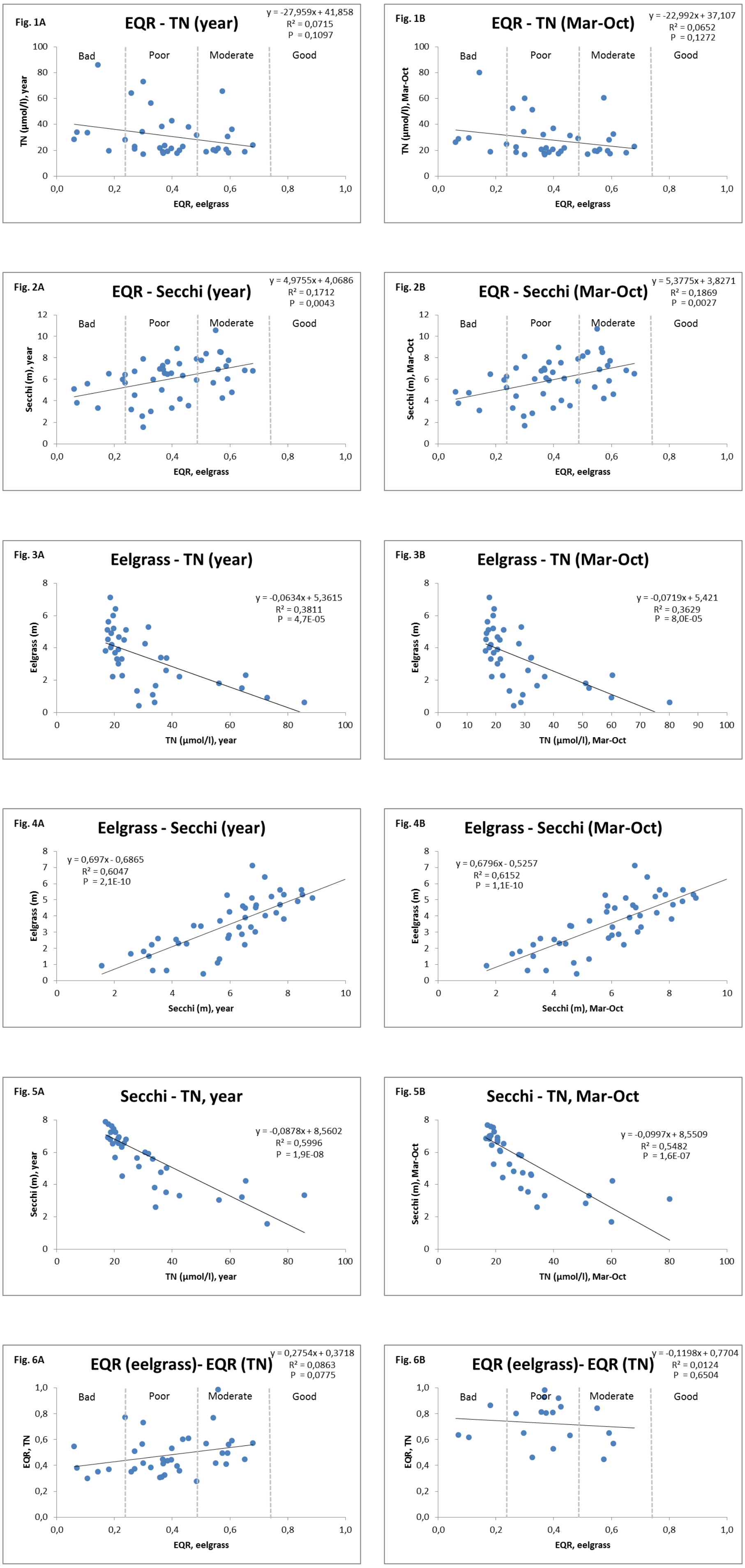
Eelgrass depth limit as a function of Secchi depth

The eelgrass depth limit from 2001-05 correlated highly significantly with Secchi depth from the same period (Figs 5.4A & B, r2<0.0001, p=0.6). The eelgrass depth limit increased with increasing Secchi depth, and the scatter around the trend line was more or less consistent for all Secchi depths.

Secchi depth as a function of TN

The Secchi depth from 2001-05 correlated highly significantly with concentrations of TN from the same period (Figs 5.5A & B, r2<0.0001, p=0.6). The Secchi depth increased with decreasing TN. However, there was a high variation of Secchi depth especially at low TN. Further, there was a steep decrease in Secchi depth with increasing TN up to 30-40 µmol l^{-1}as opposed to a very weak response in Secchi depth at higher TN levels. TN levels of 30-40 µmol l^{-1} thus seem to be a kind of N-response threshold across Danish water bodies at least for the period 2001-2005 just as found for eelgrass depth limit. Therefore, the relation between Secchi depth and TN would be better described by a non-linear function or by a linear function on each side of the 'threshold' value.

Figure 5.1. Correlations between eelgrass depth limit or EQR and environmental parameters.



Discussion

The correlation between eelgrass depth limit (2001-05) and TN concentration as well as the correlation between eelgrass depth limit and Secchi depth were highly significant even though they were based on data from a large diversity of water bodies including lagoons, fjords, bays and open water. This demonstrates that both TN and Secchi depths are important parameters regulating eelgrass depth distribution. Secchi depth is a more direct measure of light availability than TN explaining why the correlation is better for Secchi depth than TN. Low TN concentration limits the growth of plankton and thereby influence the water transparency, but water transparency is also impacted by suspended particulate inorganic and dead organic matter as well as by dissolved organic matter.

TN concentration and Secchi depth could explain around 40 % and 60 %, respectively, of the variation of eelgrass depth limit compared to 55 % and 61 %, respectively, of the variation in a study of eelgrass distribution in Danish coastal water from 1985-1991 (Nielsen et al. 2002). Both datasets represent Danish monitoring data from periods of comparable length. A comparison indicates the role of transparency of water to be more or less unchanged, whereas there may have been some uncoupling between N concentration and eelgrass depth from 1985-1991 to 2001-2005. A similar finding was reported in a previous analysis of Danish monitoring data (Carstensen & Krause-Jensen 2009).

For the 2001-2005 data, in contrast to the 1985-1991 data, the response of eelgrass depth limit and Secchi depth to increasing TN levels of at 30-40 µmol N l^{-1}. This could indicate that water bodies with long time exposure to high concentrations of TN may have a different response pattern e.g. due to fundamental changes in the system. As an example this could be former vegetated areas turned into areas with mostly bare sediment and high turbidity. In such systems there will be a lag period between reduction in the concentration of TN and improvement of the eelgrass depth limit, and the systems will most likely not recover as long as the concentration of TN is above 30-40 µmol l^{-1}.

The eelgrass depth limit EQR does not correlate well with TN or Secchi depth. The EQR represents a time component (depth limit in 2001-05 compared to reference condition) and a location component (the different water bodies) whereas TN and Secchi depth only represent a location component, which probably could contribute to the weak correlation. Thus, if depth limits of all water bodies showed the same relative deviance from reference condition, all water bodies would have similar EQR in spite of different actual levels of TN or Secchi depth. It would therefore be interesting to correlate eelgrass depth limit EQR with TN EQR and Secchi EQR in order to also include the temporal aspect in the TN and Secchi depth variables. This exercise demand access to reference TN and reference Secchi depths, which we only could provide for TN in the form of reference TN levels modelled on the basis of reference eelgrass depth limits, using a general relationship between eelgrass depth limit and TN (TN= EXP((LN (eelgrass depth limit) - 6.039)/-0.755). However, the correlation between eelgrass EQR and TN EQR was not much better than the correlation between eelgrass EQR and TN despite the auto correlation between eelgrass EQR and TN EQR (Figs 5.6A, r2=0.1, p=0.1). To avoid the auto correlation a TN EQR was also established using reference TN estimated by a site specific N-model (Carstensen et al.

2008). As expected the correlation between eelgrass EQR and this TN EQR was even worse (Fig. 5.6B, r2=0.01, p=0.7).

The large variability in the relationships described above indicate that eelgrass depth limit is regulated by many factors that besides the recent TN concentration and the Secchi depth. Factors such as physical disturbance, anoxic events (related to eutrophication), disease (such as the wasting disease in the 1930's), and possibly hazardous substances and increasing temperature can also play a role. Long-term exposure to eutrophication can also fundamentally change ecosystems resulting in severe time lacks and non-linear responses to oligotrophication. Structural changes of an ecosystem, such as vegetated areas becoming bare seafloors resulting in reduced water clarity, may have more impact on eelgrass depth distribution than recent TN levels, and may even to a certain degree uncouple the expected correlation between eelgrass depth limit and TN concentration.

The importance of other factors than TN concentration and Secchi depth for the depth distribution of eelgrass is also illustrated by the scatter around the trend line plotting eelgrass depth limit as a function of TN (Fig. 5.3A & B). Especially at low TN the variation of eelgrass depth is very high which can be explained by:

- Even at low TN the production of microalgae can be high, reducing the light availability of bottom plants, if the supply of N keeps up with the consumption. Also, other light attenuating components such as suspended particles and dissolved organic matter can be important even at low TN.

- The system response to low TN concentration varies according to the residence time and the importance of the internal nutrient supply, which both differs a lot between water bodies.

- The eelgrass depth limit may be low despite low TN if the light availability is controlled by resuspension more than by plankton as it is the case in shallow wind exposed water bodies.

- Physical disturbance can dramatically change eelgrass distributions e.g. if eelgrass is covered in sediments or removed due to erosion during a storm event.

Even though TN concentrations and Secchi depth are main factors regulating eelgrass their relationship to eelgrass depth limits are thus not necessarily linear and responses may vary between water bodies.

5.2 Macroalgal on reefs in more open waters and algal vegetation in fjords and coastal areas

In Denmark a set of algal metrics (indicators) and methodologies have been developed and used to describe reference conditions and the present ecological status.

Empirical models have been developed for NATURA-2000 reef sites in the open part of Kattegat describing 'total vegetation cover' and 'cumulative vegetation cover' as a function of locality, solar radiation, depth, grassing pressure of sea-urchin, and total load of nutrient to Kattegat (Dahl & Carstensen, 2008). Both models are statistically well founded. Lacking historic information on the vegetation cover in Danish waters the two models have been used to describe reference conditions and other scenarios of vegetation cover at different estimates of nitrogen loads to Kattegat, as seen in the example in Fig. 5.7. The reefs sites are located in open waters outside the coastal area covered by the Water Framework Directive.

Empirical modelling on algal datasets collected on hard stable substrate in fjords and shallow coastal areas of Denmark has also been done recently (Carstensen et al., 2008). More or less all datasets collected as part of the Danish national monitoring program in 2001, 2003 and 2005 have been included in the work. Six different indicators were tested and important structuring factors identified and quantified. Common for all indicators are that data has been normalised for differences in sampling depth, spatial variation within a water body and time within the summer period by an 'underlying model'. The resulting algal indicators are expressed as an average value and variance representing 7 m water depth in each selected water body. Each of the six indicators and the factors that significantly structured the vegetation in the analysed dataset are shown in Table 5.2.

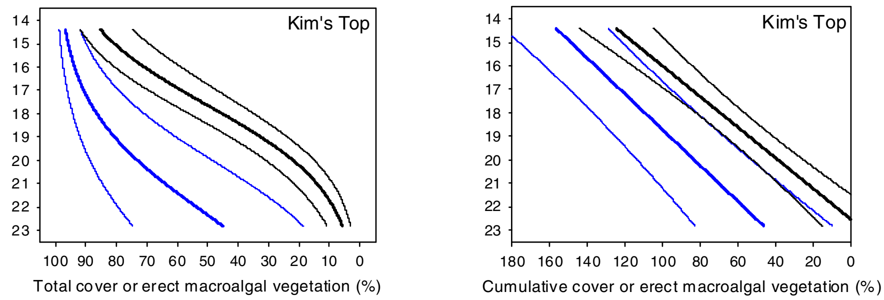


Figure 5.7. Total cover (left) and cumulative cover (right) of macro-phytes at different depth and at different nutrient load scenarios at the reef Kim's Top in the central part of Kattegat. The thick blue line describes a reference load scenario with 10.000 tons from rivers and point sources in January-June and the thick black line describes an average load scenario equal to the period 1999-2007 on 48.000 tons in the same six month. The thin lines describe the upper and lower 95% confidence intervals on the estimated covers. From Dahl & Carstensen (2008).

Table 5.2. Macroalgal indicators and factors significantly structuring each indicator, as well as the overall model correlations. TN =Total Nitrogen TN*salinity means that the effect of TN was dependent on salinity. The arrows indicate positive (up) or negative (down) effects of TN and salinity (from Carstensen et al. 2008).

Variable	TN	Salinity	TN * Salinity	R^2
Total algal cover		↑	↓	0.68
Cumulated algal cover		↑	↓	0.70
Cumulated cover of per-annual species	↓	↑		0.71
Cumulated cover of opportunistic species			↓	0.69
Fraction of opportunistic algal species	↑		↓	0.63
Number of per-annual species		↑	↓	0.79

All macroalgal variables responded significantly ($p<0.05$) to a combination of changes in total nitrogen and to changes in salinity which emphasises the need for setting different targets depending on salinity. The strongest responses to changes in nitrogen concentration and the least variability were found for the indicators 'total algal cover', 'number of late-successional species', and 'fraction of opportunists' in less saline waters. The number of late successional species is regarded as an indicator of biodiversity.

As was the case with reef vegetation in open waters no reference data is available for macroalgal vegetation in coastal Danish waters. Reference conditions for each algal indicators and ecological status class boundaries were established for all the macroalgal variables in a large number of water bodies, considerably smaller than prescribed in the Water Framework Directive. The boundaries were established based on estimates on pristine load scenarios and site-specific relations between load and concentrations in the recipient water bodies. Fig. 5.8 gives an example of all indicators from the north-western part of Limfjorden, a water body with excellent datasets of both hydrography and algal stations.

All though the models for coastal waters overall had a good correlation with nitrogen and salinity, its availability as assessment tool with regard to the Water Framework Directive was restricted to a number of fjord areas. In some fjord areas the data input was probably to scare. In open waters the chosen depth of 7 meter in the under laying depth model was not optimal as there were hardly any difference and large overlap in the estimated class boundaries for the most suitable indicator 'total cover'.

The methodology of linking macroalgal covers to water chemistry used in Danish waters has also been tested on a dataset from Finland. Krause-Jensen et al. (2009) found that Finnish and Danish coastal monitoring data of cumulative cover (sum of all species-specific cover) had a similar functional relationship to Secchi depths. In this study it was hypothesized that a common functional relationship of total cover to Secchi depths can be obtained across differences in the national monitoring programs.

Recent studies on a Baltic wide dataset (Skov et al. in prep.) have documented significant negative effects of eutrophication on total macroalgal cover (TC) across the open Norwegian North Sea to the inner Baltic See. In the entire region, Secchi depth declined in response to increasing TN, and in more brackish areas also in response to increasing TP. In general there was a large scatter in the relationships established between eutrophication variable and vegetation cover. A subset of data from the open part of Kattegat, Skagerrak, and a small part of the Norwegian North Sea coast were most robust, in the sense that "country" (most likely representing different sampling methods) was not found to be significant in the analysis. It is likely that total cover might be a common matrix for this area in years to come. Sampling of 'total cover' is relative new in the Swedish and Norwegian monitoring program and boundary setting have to be done on a basin wide scale based on empirical models as described above.

Conclusion: At present there is not a joint Danish assessment tool combining depth distribution of eelgrass, cover of macrophytes, and number of perannual species. However each of those single elements have been tested against nitrogen concentration or nitrogen load which is regarded as the most important anthropogenic pressure in inner Danish waters.

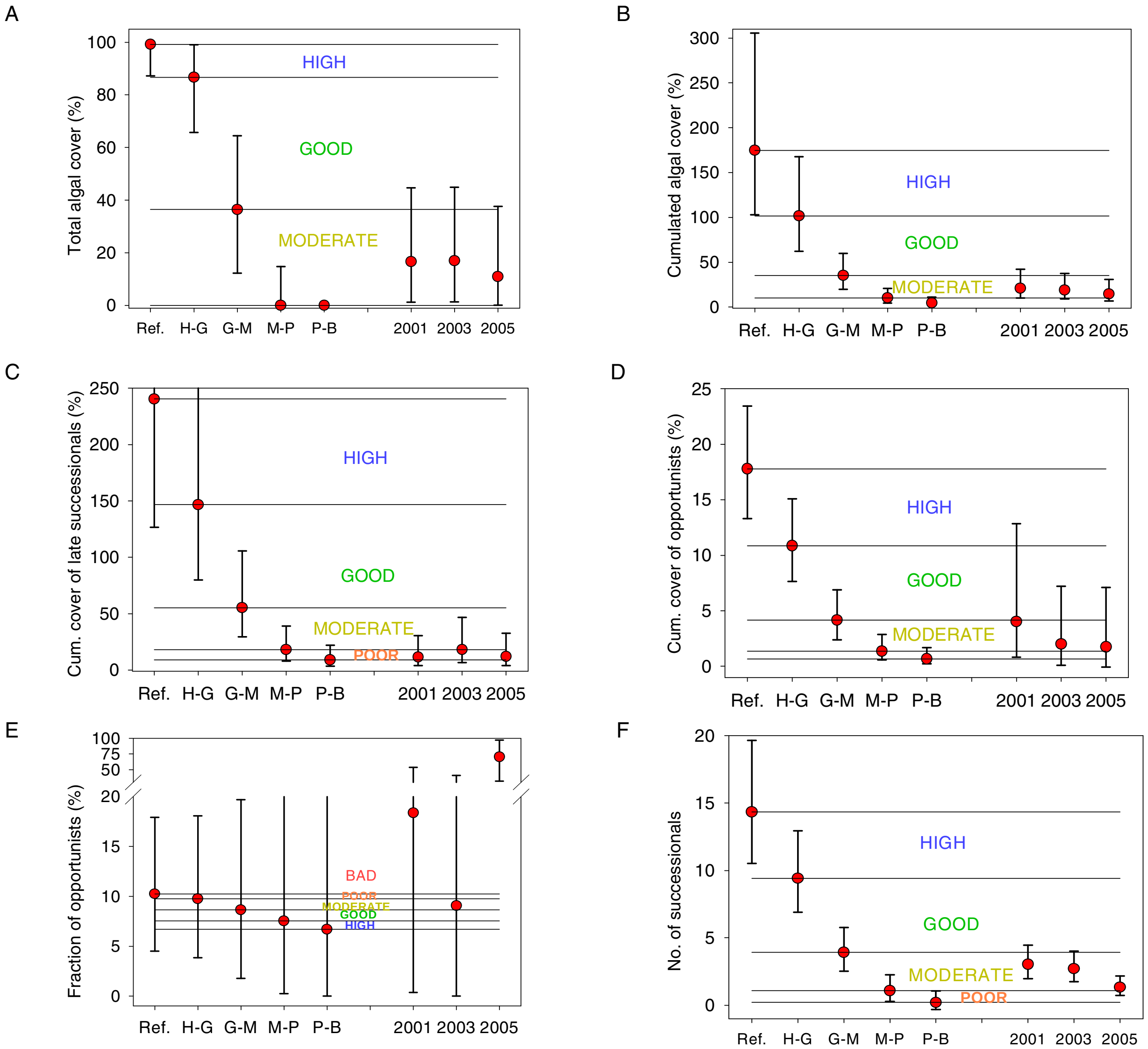


Figure 5.8. Reference levels, class borders and actual levels of various algal variables in Limfjorden west of Mors.) Total algal cover (A), Cumulated algal cover (B), Cumulated cover of late-successionals (C), Cumulated cover of opportunists (D), fraction of opportunists (E) and number of late-successional algal species (F). Algal variables are modelled for a water depth of 7 m.

5.2.1 Intercalibration: Denmark-Sweden Baltic GIG

This area covers the Danish typology OW3 and the Swedish typology 7 (Fig. 5.9).

It was not possible to find a suitable dataset for intercalibration between Denmark and Sweden in the Baltic waters south of the bridge.

The Danish coastal macroalgal assessment tool is not working properly in neither Køge nor Hjelm Bays in the present setup. The tool is focused on an output for fjords with less water transparency. Unfortunately there were no available resources for optimizing and testing the assessment tool for open coastal stations.

Depth distribution of selected algal species is used as a Swedish BQE. Danish macroalgal data are collected and reported within 2 m depth intervals introducing a huge uncertainty but substrate limitation is often the limiting factor for algal vegetation in coastal Danish waters as well. The Swedish methodology was not tested on Danish data.

Eelgrass is registered in the Swedish typology but data on depth distribution was not collected. Depth distribution is used in the Danish assessment tool for eelgrass. No class boundaries have been established in Swedish waters.

It was not possible to find a common metric for intercalibration (option 2).

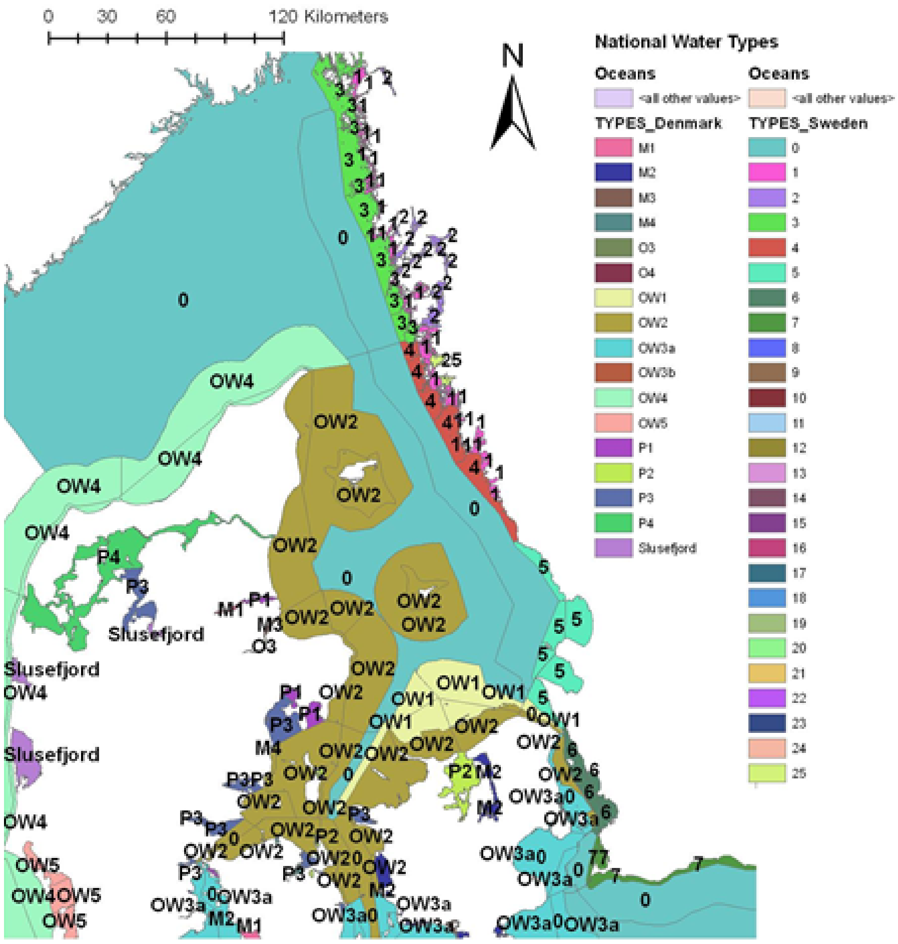


Figure 5.9. Distribution of the Danish typology OW3 and the Swedish typology 7.

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DANISH CONTRIBUTION TO THE EU WATER FRAMEWORK DIRECTIVE INTERCALIBRATION PHASE 2

This report presents the Danish contributions to the European Water Framework Directive intercalibration phase 2. The project was initiated and financed by the Danish Nature Agency and focused on development of phytoplankton indicators based on biovolume and carbon biomass, further development and adjustment of the Danish benthic fauna index, and evaluating the possibilities of intercalibrating indicators of benthic vegetation. The results of this project have been included in the Baltic GIG and NEA GIG Milestone 6 reports.

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