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**Colourful coexistence : a new solution to the plankton paradox**

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# Appendix A

## Sampling stations

During the summers of 1986-1988, 30 lakes in Canada and 3 lakes in New Zealand were sampled, covering a wide range in background turbidities and water-column depths (Pick 1991). For each lake, 8-12 samples were taken from 2 m depth using a Van Dorn sampler. These samples were mixed. During the summers of 1994 and 1995, 13 lakes in Hungary, 12 lakes in Italy and 2 lakes in Nepal were sampled (Vörös *et al.* 1998). In the deep lakes, the first 20 m of the water column was sampled with an integrating sampler. In the shallow lakes, ponds and reservoirs the whole water column was sampled by a Van Dorn sampler using an interval of 1 m, and these samples were mixed. From 12 to 19 July 2004, 9 stations in the Baltic Sea (from 59.1°N to 60.0°N and from 22.2°E to 26.2°E) were sampled from the research vessel Aranda on Cruise Cyano-04 08/2004. Water samples were taken with a Rosette sampler from 0 to 30 m depth using a sample interval of 3 m. Temperature was measured using the Seabird 911 plus CTD sonde. From 5 to 11 October 2005, Station ALOHA (23.4°N, 158°W) of the Hawaiian Ocean Time series (HOT) in the North Subtropical Pacific Ocean was sampled from the research vessel Kilo Moana on cruise number 174. Water samples were taken from 12 depths within the upper 200 m with a SeaBird (Model SBE-09) CTD Rosette system. An overview of all 70 sampling stations is given in Table A1.

## Measurement of background turbidity

To calculate the underwater light field, the model uses the background turbidity at the reference wavelength of 484 nm,  $K_{BC}(484)$ , as input parameter (Equation 4.4). We determined  $K_{BC}(484)$  spectrophotometrically, as the sum of the light absorption by gilvin,  $K_{GL}(484)$ , and the light absorption by tripton,  $K_{TRIP}(484)$ .

*Absorption by gilvin:* Dissolved organic matter is known as 'gilvin' in the optics literature. To determine light absorption by gilvin, water samples were filtered through 0.2 µm cellulose acetate filters (Schleicher and Schuell). Absorption spectra of the filtrate were measured by a Lambda 800 UV/VIS spectrophotometer (Perkin-Elmer, Wellesley, MA, USA) using a 5 cm quartz cuvet, with milli-Q water as reference (Simis *et al.* 2005). The parameter  $K_{GL}(484)$  is the light absorption by gilvin measured at 484 nm.

*Absorption by tripton:* Tripton refers to inanimate suspended particles in the water column. Absorption spectra of suspended matter were determined on GF/F filters using the filterpad method (Yentsch 1962; Cleveland & Weidemann 1993; Simis *et al.* 2005). The spectra were measured with a Lambda 800 UV/VIS spectrophotometer (Perkin-Elmer, Wellesley, MA, USA) equipped with a 150-mm integrating sphere (Labsphere, North Sotton, NH, USA). For the correction of path length amplification the method of Cleveland and Weidemann (1993) was used. First, the absorption spectrum of the loaded filter, obtained after filtration of

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the water sample, was measured. This includes all seston (phytoplankton plus tripton). As a next step, the absorption spectrum of tripton on the filter was measured, after bleaching of phytoplankton pigments by boiling ethanol. The parameter  $K_{TRIP}(484)$  is the light absorption by tripton measured at 484 nm.

### An algorithm to calculate background turbidity

Ideally, one would like to determine the background turbidity from direct measurements of the light absorption by gilvin and tripton, as described above. However, for several sampling stations we did not have data on the absorption by gilvin and tripton. Therefore, we developed a simple algorithm to calculate the background turbidity from the total light attenuation coefficient and the chlorophyll concentration in the water column as described below.

#### Partitioning of the total light attenuation

The total light attenuation,  $K_D$ , in natural waters is governed by light attenuation by gilvin and tripton,  $K_{BG}$ , attenuation by water itself,  $K_W$ , and attenuation by phytoplankton,  $K_{PHYT}$  (Kirk 1994). Hence, the total light attenuation at the reference wavelength of 484 nm can be partitioned as follows:

$$K_D(484) = K_{BG}(484) + K_W(484) + K_{PHYT}(484) \quad (A1)$$

Accordingly,  $K_{BG}(484)$  can be calculated if the values of the other attenuation coefficients in Equation A1 are known. The total light attenuation coefficient at 484 nm,  $K_D(484)$ , was estimated from the attenuation coefficient of photosynthetic active radiation,  $K_D(\text{PAR})$ , using the empirical relation (Balogh *et al.* 2000):

$$^{10}\log[K_D(484)] = 1.1353 \ ^{10}\log[K_D(\text{PAR})] + 0.2023 \quad (A2)$$

where  $K_D(\text{PAR})$  was estimated from vertical light profiles (PAR range, 400-700 nm), measured with a Licor Li-185 quantum sensor for the Baltic Sea and the lakes in Hungary, Italy and Nepal and with a Licor Li-190 quantum sensor for the lakes in Canada and New Zealand. Light attenuation by pure water at 484 nm is known, i.e.,  $K_W(484) = 0.0136 \text{ m}^{-1}$  (Pope & Fry 1997). Light attenuation by phytoplankton,  $K_{PHYT}(484)$ , was calculated from chlorophyll *a* concentrations, as described below.

#### Absorption by phytoplankton at 484 nm

We established a relationship between  $K_{PHYT}(484)$  and the chlorophyll concentration. For this purpose, samples from 10 sampling stations in the Baltic Sea, at 11 different depths per sampling station, were each split into two subsamples. One set of subsamples was used for chlorophyll analysis while the other set of subsamples was used to determine the phytoplankton absorption spectra. Chlorophyll *a* concentrations were measured

spectrophotometrically after hot ethanol extraction of phytoplankton collected on Whatman GF/F filters (Nusch 1980). Light absorption spectra of the phytoplankton communities were obtained from the filterpad method, as the difference between the absorption spectrum of seston (phytoplankton plus tripton) and the absorption spectrum of tripton. The results show a strong relationship between the phytoplankton light absorption at 484 nm and the chlorophyll *a* concentration (Figure A1a):

$$K_{PHT}(484) = 0.0368 [\text{Chl}] \quad (\text{A3})$$

where [Chl] is the chlorophyll *a* concentration in  $\mu\text{g Chl L}^{-1}$  (linear regression forced through the origin:  $R^2 = 0.93$ ,  $n=110$ ,  $p<0.0001$ ). Equation A3 was used to calculate  $K_{PHT}(484)$  from the chlorophyll *a* concentrations for all sampling stations.

#### *Calibration of the algorithm*

The background turbidity,  $K_{BG}(484)$ , can now be calculated from Equations A1-A3. To test this, we compared the predicted background turbidity (Equations B1-B3) with the measured background turbidity. For this purpose, we applied Equation A1-A3 to an independent data set consisting of 5 Dutch lakes (Lake Loosdrecht, Lake Proost, Lake Groote Moost, Lake t'Elfde, Lake IJsselmeer), 9 sampling stations in the Baltic Sea, and 2 stations near station ALOHA (Pacific Ocean, Hawaii). At these sites we also measured the background turbidity following the described algorithm. This showed a close correspondence between the predicted and measured background turbidity (Figure A1b):

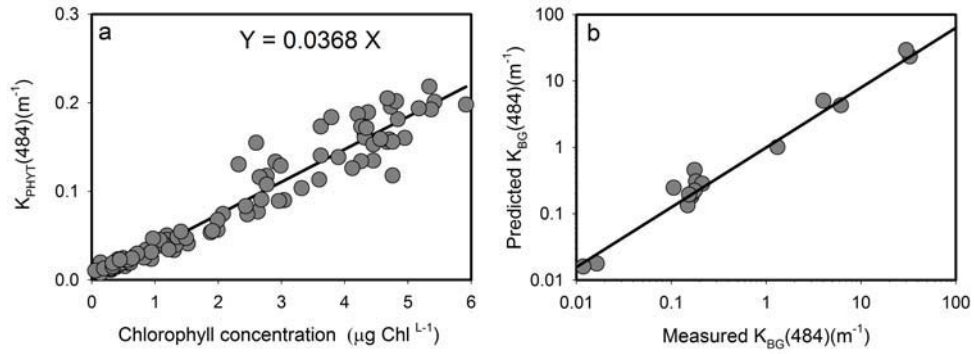
$$^{10}\log[K_{BG,pred}(484)] = 0.9006 \ ^{10}\log[K_{BG,meas}(484)] \quad (\text{A4})$$

based on linear regression forced through the origin, after log-transformation of the data ( $R_2=0.98$ ,  $n=16$ ,  $p<0.0001$ ). The factor 0.9006 in Equation A4 was incorporated as correction factor in the algorithm to improve our predictions. Hence, combining the information in Equations A1-A4, the following algorithm was obtained to predict the background turbidity at 484 nm from the light attenuation coefficient and chlorophyll *a* concentration:

$$K_{BG}(484) = \left[ 1.593 [K_D(PAR)]^{1.135} - 0.0136 - 0.0368 [\text{Chl}] \right]^{1.11} \quad (\text{A5})$$

We applied this semi-empirical algorithm to calculate the background turbidity at all 70 sampling stations. Furthermore, we suggest that the algorithm may also find application in other studies.

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**Figure A1** (a) Light attenuation coefficient of phytoplankton at 484 nm,  $K_{\text{PHYT}}(484)$ , as function of the chlorophyll *a* concentration. (b) Background turbidity predicted from Equations A1-A3 against measured background turbidity. Data points represent samples from 5 Dutch lakes (Lake Loosdrecht, Lake Proost, Lake Groote Moost, Lake t'Elfde, Lake IJsselmeer), 9 sampling stations in the Baltic Sea, and 2 sampling stations near station ALOHA (Pacific Ocean, Hawaii).

**Table A1** Sampling stations and some of their characteristics

Sampling stations	Area (km <sup>2</sup> )	Average depth (m)	Sampling depth (m)	K <sub>BG</sub> (484) (m <sup>-1</sup> )	Red picos (%)
<b>Hungary, lakes</b>					
L. Balaton (Fűzfő Basin)	596	3.2	0 – 2	1.65	73
L. Balaton (Tihany basin)	596	3.2	0 - 3.7*	2.73	57
L. Balaton (Zánka basin)	596	3.2	0 – 2	1.94	63
L. Balaton (Szigliget basin)	596	3.2	0 – 2	1.49	27
L. Balaton (Keszthely basin)	596	3.2	0 - 2.3*	2.17	73
L. Balaton (Zala river)	-	-	-	3.03	6
Kis-Balaton (upper res.)	18	1	0 – 1*	3.82	0
Kis-Balaton (lower res.)	16	0.8	0 - 0.8*	4.66	0
Marcali reservoir	4	1.8	0 - 1.8*	3.48	0
Monostorapáti reservoir	0.3	2	0 – 2*	6.00	4
L. Pécsi	0.75	3.3	0 - 3.3*	1.41	78
L. Herman Otto	0.29	1	0 – 1*	3.95	32
Deseda reservoir	2.2	2.9	0 - 2.9*	5.90	2
<b>Italy, lakes</b>					
L. Como	146	154	0 – 20*	0.28	98
L. Maggiore	212	177	0 – 20*	0.43	96
L. Garda	368	133	0 – 20*	0.22	98
L. Iseo	62	123	0 – 20*	0.29	99
L. Orta	18	72	0 – 20*	0.46	100
L. Mergozzo	1.8	45	0 – 20*	0.25	99
L. Varese	15	11	0 – 11*	0.69	54
L. Candia	1.3	5.9	0 - 5.9*	0.49	50
L. Paione Superiore	0.014	5.1	0 - 5.1*	0.35	100
L. Paione Inferiore	0.014	7.3	0 - 7.3*	0.12	100
L. Azzuro	0.003	2	0 – 2*	0.30	100
L. Devero	1	20	0 – 20*	0.35	100
<b>Nepal, lakes</b>					
L. Piramide Superiore	0.6	8.2	0 - 8.2*	0.21	100
L. Piramide Inferiore	1.7	14.8	0 - 14.8*	0.12	100
<b>New Zealand, lakes</b>					
Okareka	3.5	12	2	0.30	55
Tarawera	41	50	2	0.36	100
Rotorua	80	6.8	2	0.80	60
<b>Ontario, lakes</b>					
Superior	81900	145	2	0.19	100
Erie (east)	6150	27	2	0.42	100
Erie (central)	15390	18	2	0.32	100
Erie (west)	3680	7.6	2	0.94	67
Ontario	19680	90	2	0.41	100

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Table A1 continued.

Bay of Quinte	257	8.3	2	2.15	11
Cherry	0.22	5.5	2	0.83	16
Triangle	0.27	4.7	2	0.59	49
Bay	1.6	11	2	0.35	100
Buller	0.31	20	2	0.46	100
Halls	5.7	?	2	0.24	88
Koshlong	4.1	10	2	0.48	68
Anstruther	6.3	13	2	0.69	19
L'Amable	1.8	23	2	0.48	100
Opeongo	22	?	2	1.21	21
St. Nora	?	?	2	0.91	52
Crawford	0.02	?	2	0.45	100
Drag	10	18	2	0.48	92
Wolf	1.2	4.8	2	0.86	35
Picard	0.76	10	2	0.48	99
Salmon	1.7	11	2	0.35	100
Bobs GB	4.8	14	2	0.41	93
Chub	0.34	8.8	2	0.85	0
Jacks	5.1	17	2	0.57	95
Bobs WB	9.4	9.5	2	0.74	66
St. George	0.10	/	2	0.66	53
Rice	100	2.4	2	1.52	18
Heart	0.18	3.7	2	2.34	0
<b>Alberta, lakes</b>					
Island	7.8	3.7	2	0.76	90
Amisk	5.2	16	2	1.11	91
<b>Baltic Sea</b>					
LL3A	$3.7 \times 10^5$	69	0 – 20*	0.86	55
CYA04_2	$3.7 \times 10^5$	75	0 – 14*	0.31	77
CYA04_3	$3.7 \times 10^5$	63	0 – 17*	0.70	69
CYA04_7	$3.7 \times 10^5$	85	0 – 14*	0.63	72
CYA04_11	$3.7 \times 10^5$	77	0 – 15*	1.17	39
CYA04_15	$3.7 \times 10^5$	69	0 – 15*	0.67	66
CYA04_20	$3.7 \times 10^5$	62	0 – 30*	0.56	51
CYA04_22	$3.7 \times 10^5$	90	0 – 20*	0.71	52
CYA04_28	$3.7 \times 10^5$	111	0 – 40*	0.52	65
<b>Pacific Ocean, Hawaii</b>					
ALOHA	N.A.	~4000	0 – 120*	0.016	100

\*Samples were integrated over the depth of the surface mixed layer