

10th Webinar

Do you know your phytoplankton?

Go deep - best practice of profiling with the FluoroProbe



Welcome



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10th Webinar

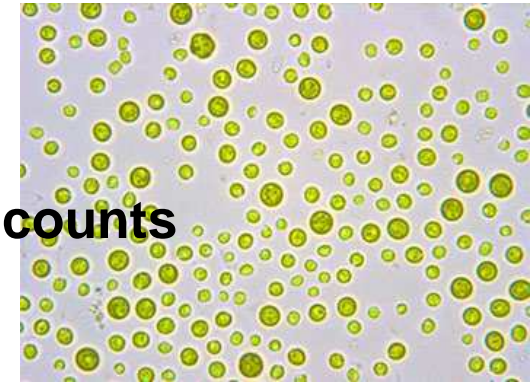
Part 1	Principle
Part 2	Performance
Part 3	Data and Software
Part 4	Calibration

Feedback
Follow up

How to measure Algae?



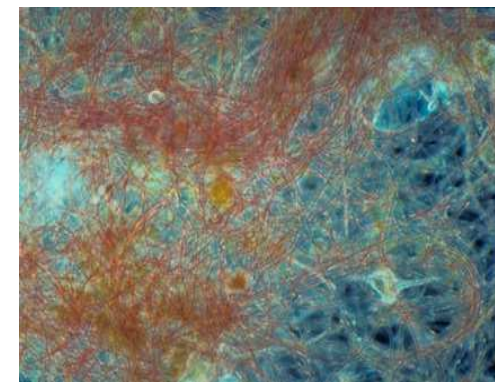
- **microscopic analysis = cell counts**
 - **slow**
 - **laborious**
 - **counting errors**



Chlorella vulgaris



- **fluorometric method = chlorophyll a**
 - **fast**
 - **simple performance**
 - **reliable**
 - ***in vivo* & ultra sensitive**

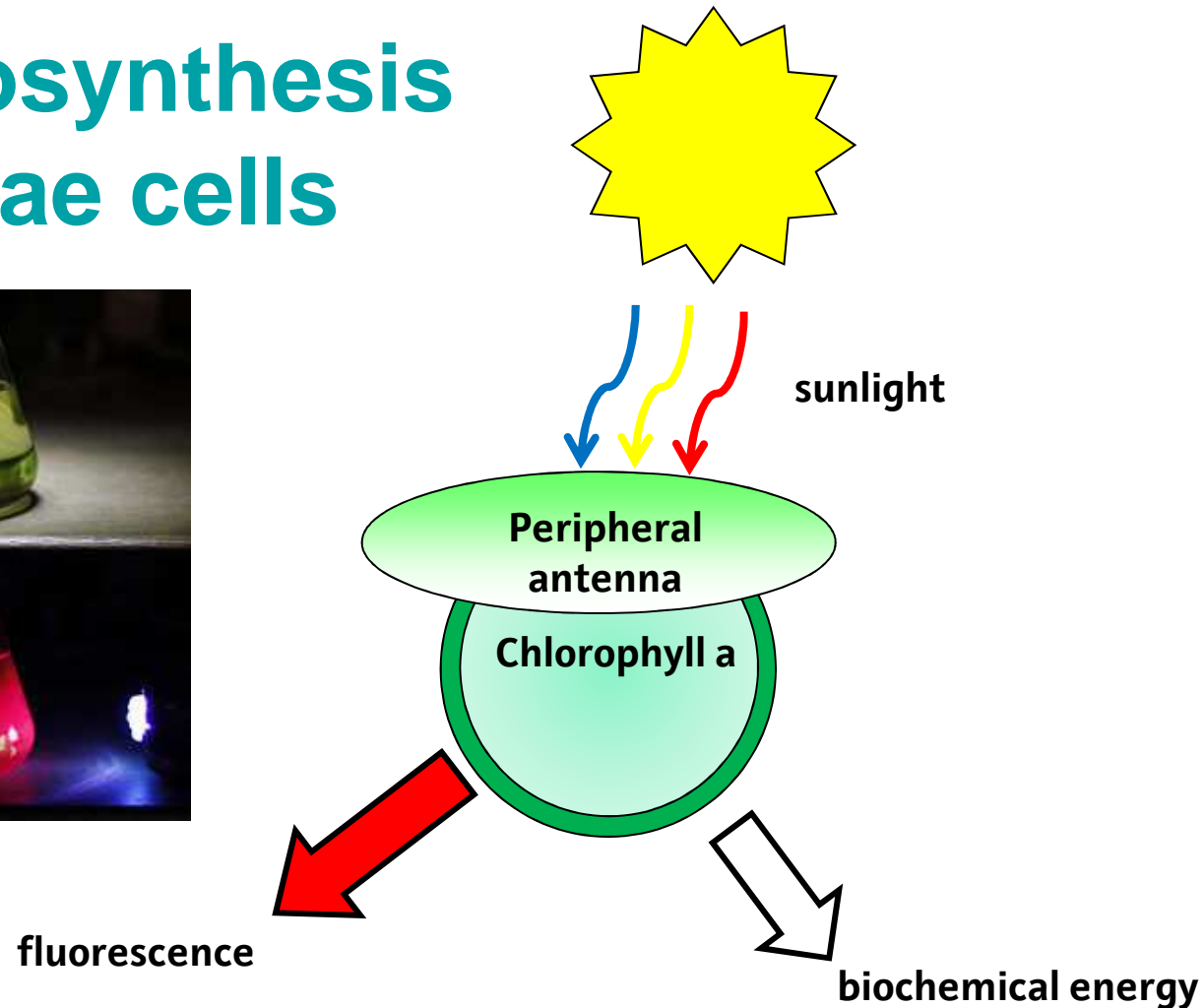


Planktothrix rubescens

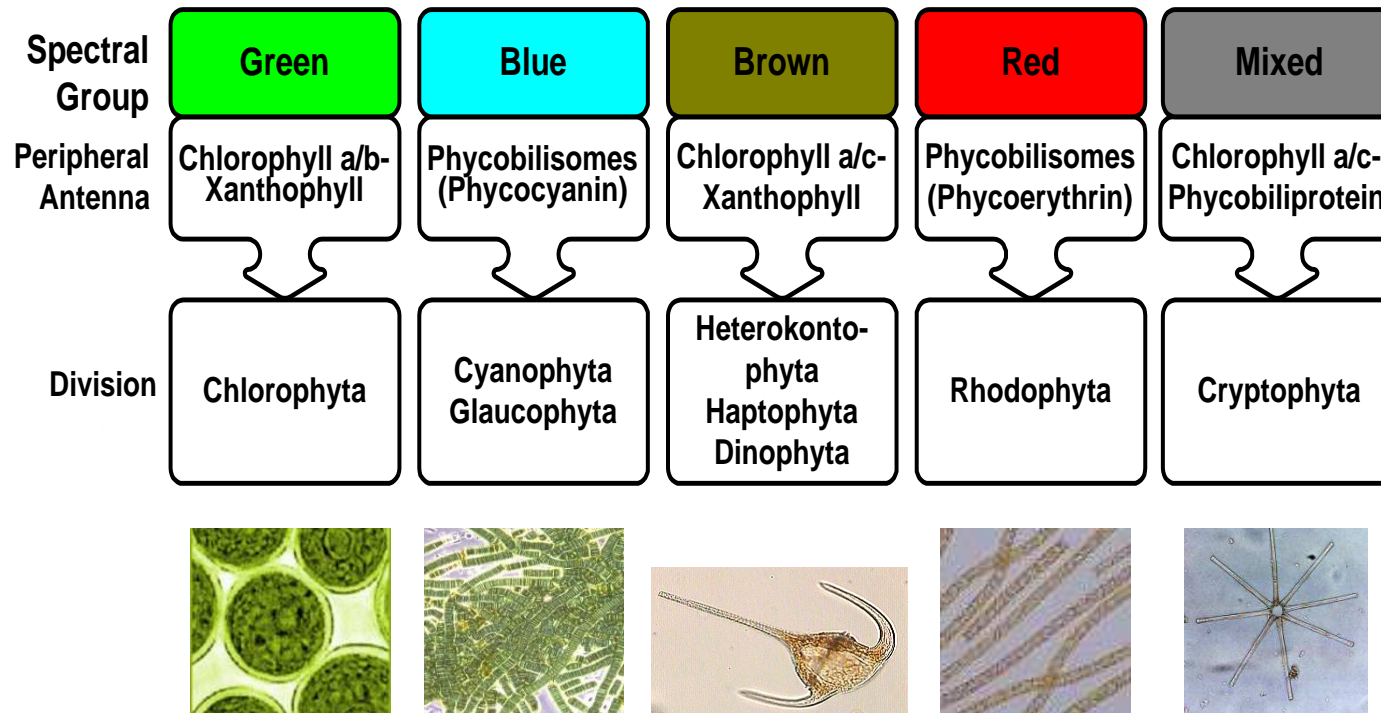
Depth Profiling of Algae Populations by Fluorometric Analysis using the bbe FluoroProbe



Photosynthesis in algae cells

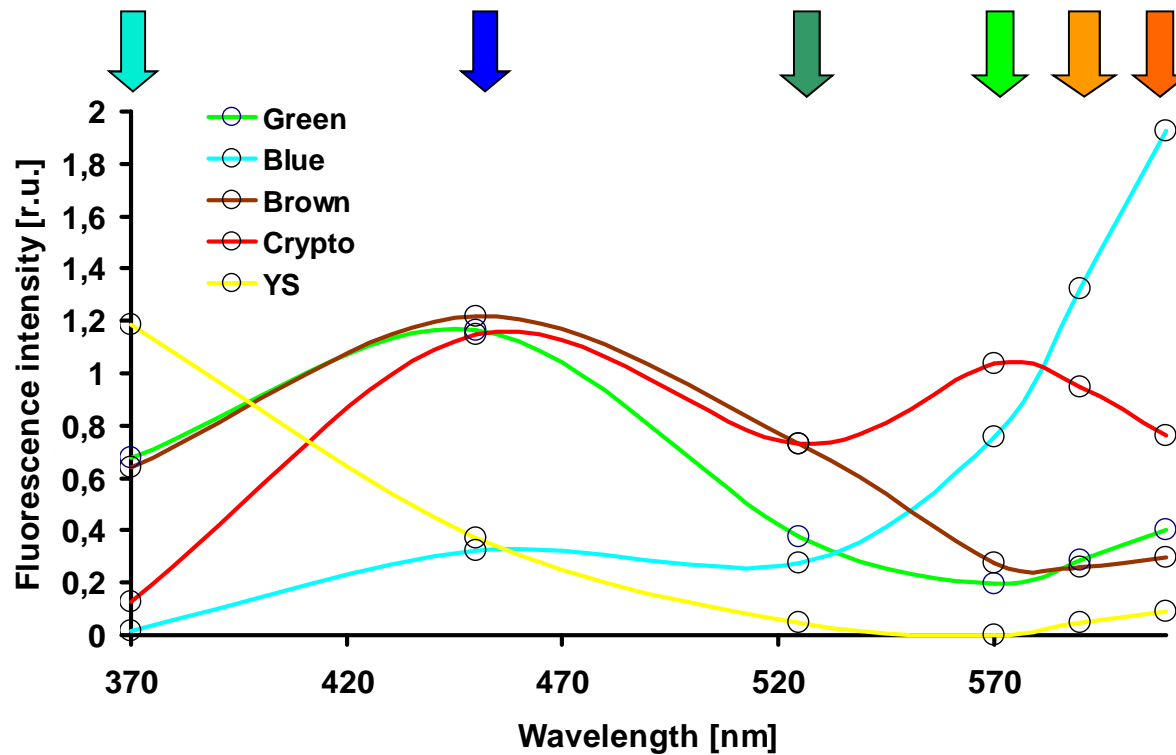


Algae classes contain different accessory pigments



...which affect the chlorophyll a fluorescence emission in characteristic pattern

Spectral chlorophyll fingerprints with the FluoroProbe

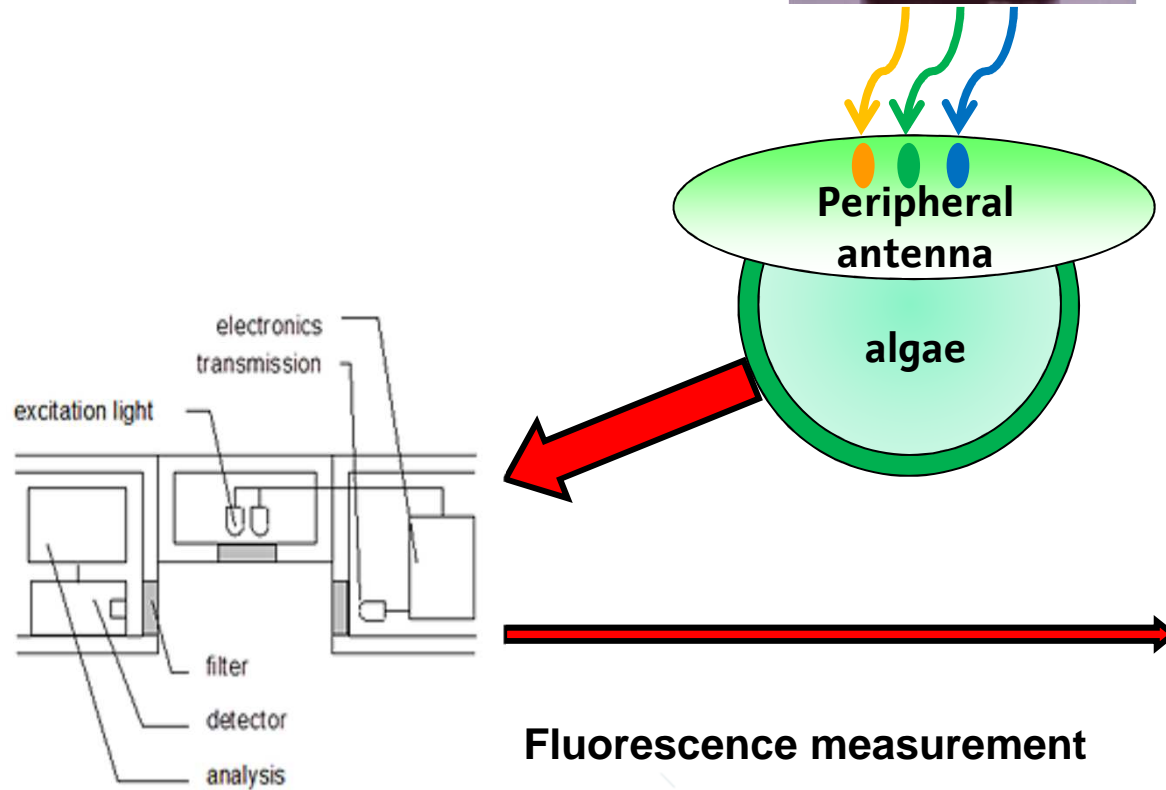


conventional fluorometer

bbe measuring principle



multicolour
excitation with LED



Charging the Probe / Mode



Charge state	Color
Full	Green
Medium	Yellow-Orange
Nearly empty	Red

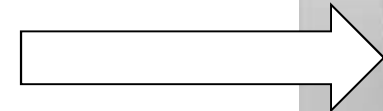
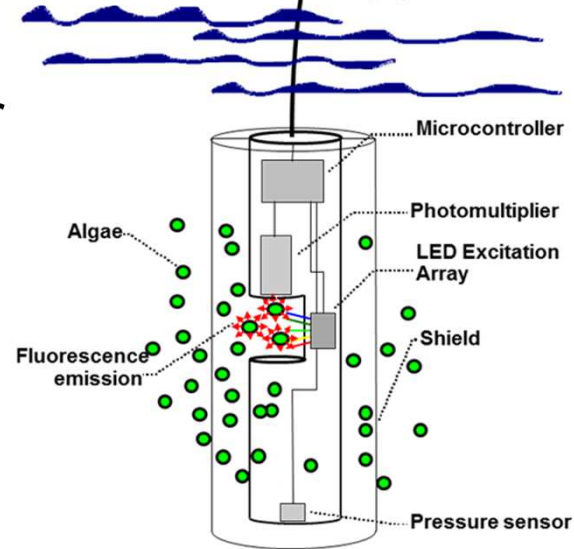


Mode	Blinking
Off	Off
Standby	Short light pulses
Measuring	Regular pulses
Charging	On

Depth measurement

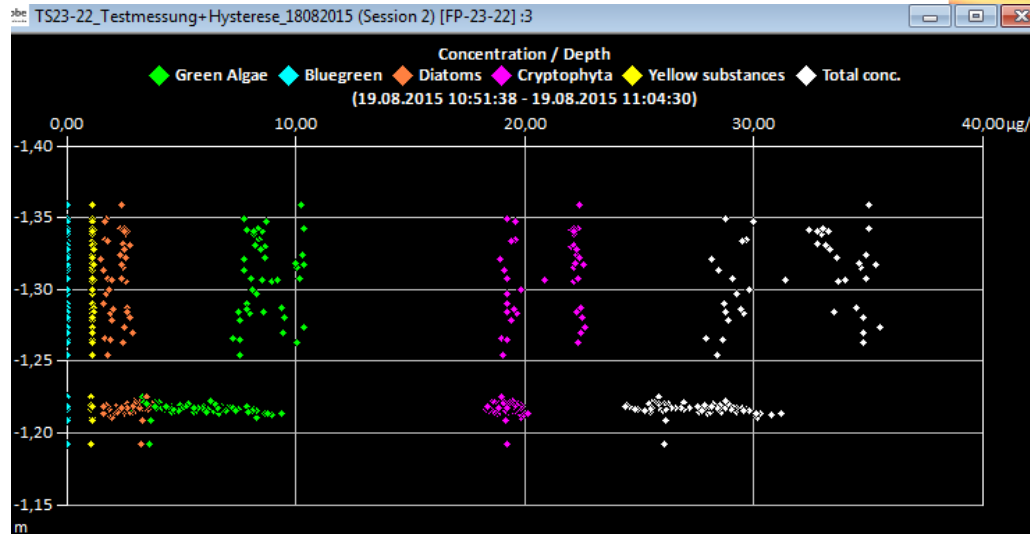


1 m = 0,1 bar



Depth profiling

- Position
- Speed
- Frequency of measurement
- Direction



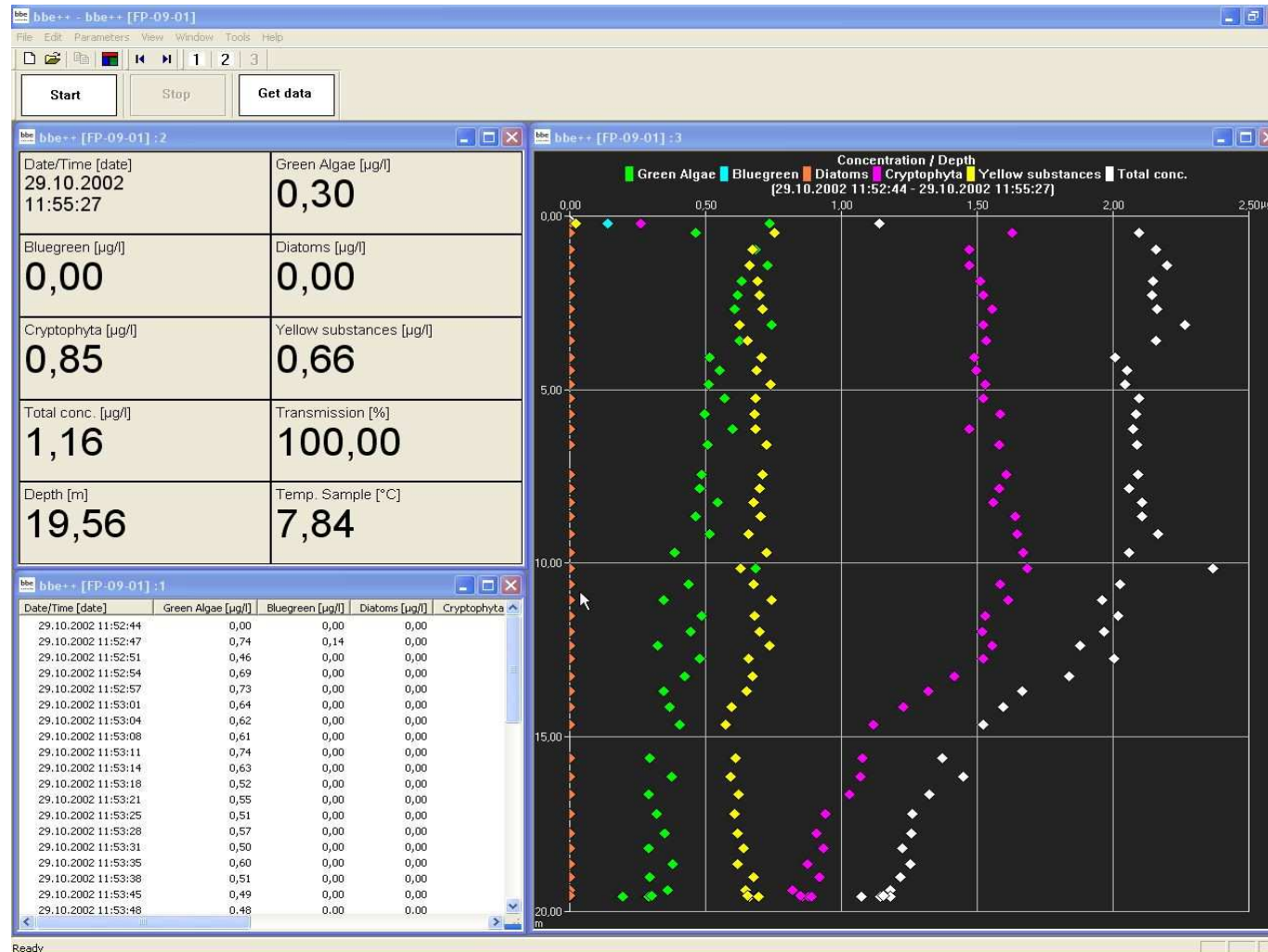
Outer Factors

- Sunlight
- Surface reflection
- Turbidity
- Biofouling

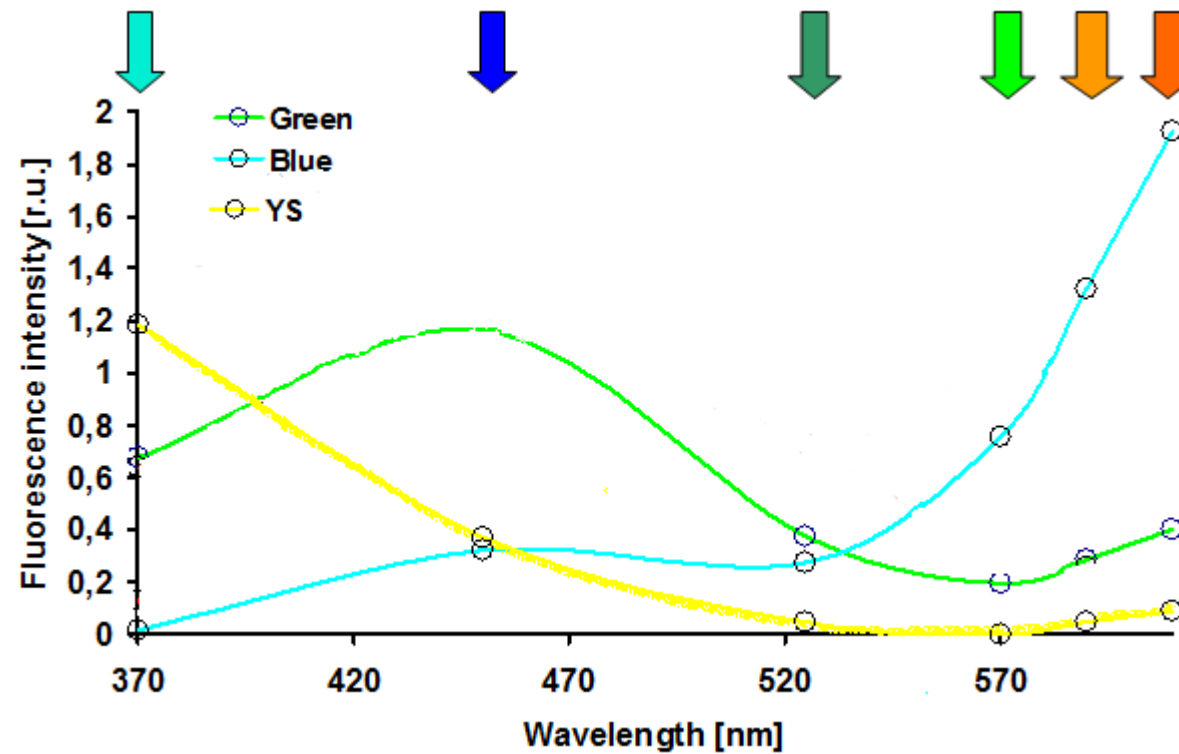
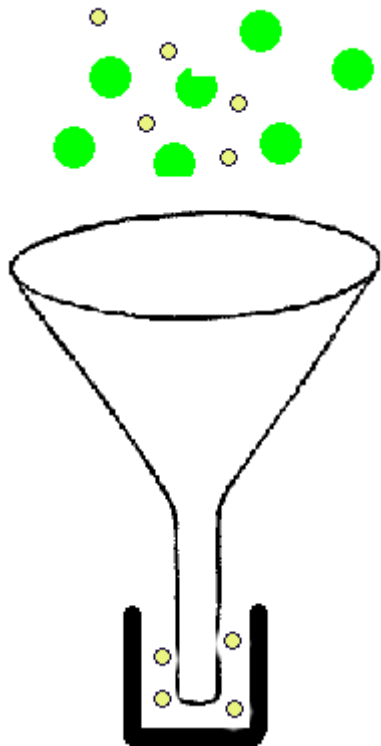


Common parameters		Fit parameters		Measurement parameters		Turbidity parameters	
Name				Value	U		
Turbidity compensation				off			
FTU polynomial factor a				0,03			
FTU polynomial factor b				-5,9174			
FTU polynomial factor c				294,16			
FTU reflection compensation LED 3 [525 nm]				0,01577			

Yellow Substances & Humic Acids



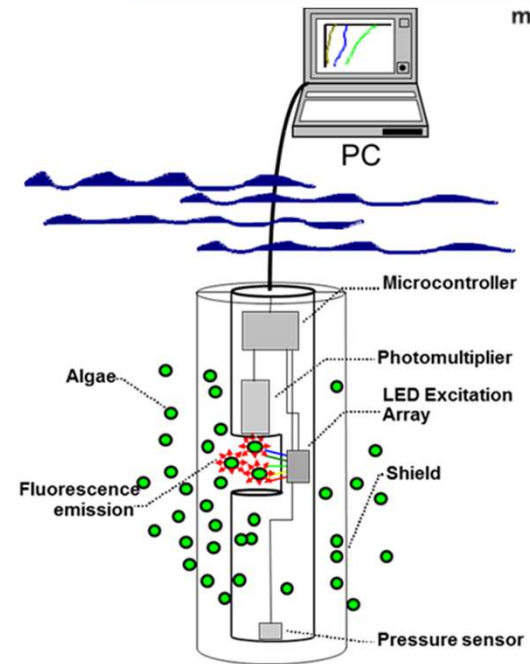
Compensation for yellow substances = 2 methods



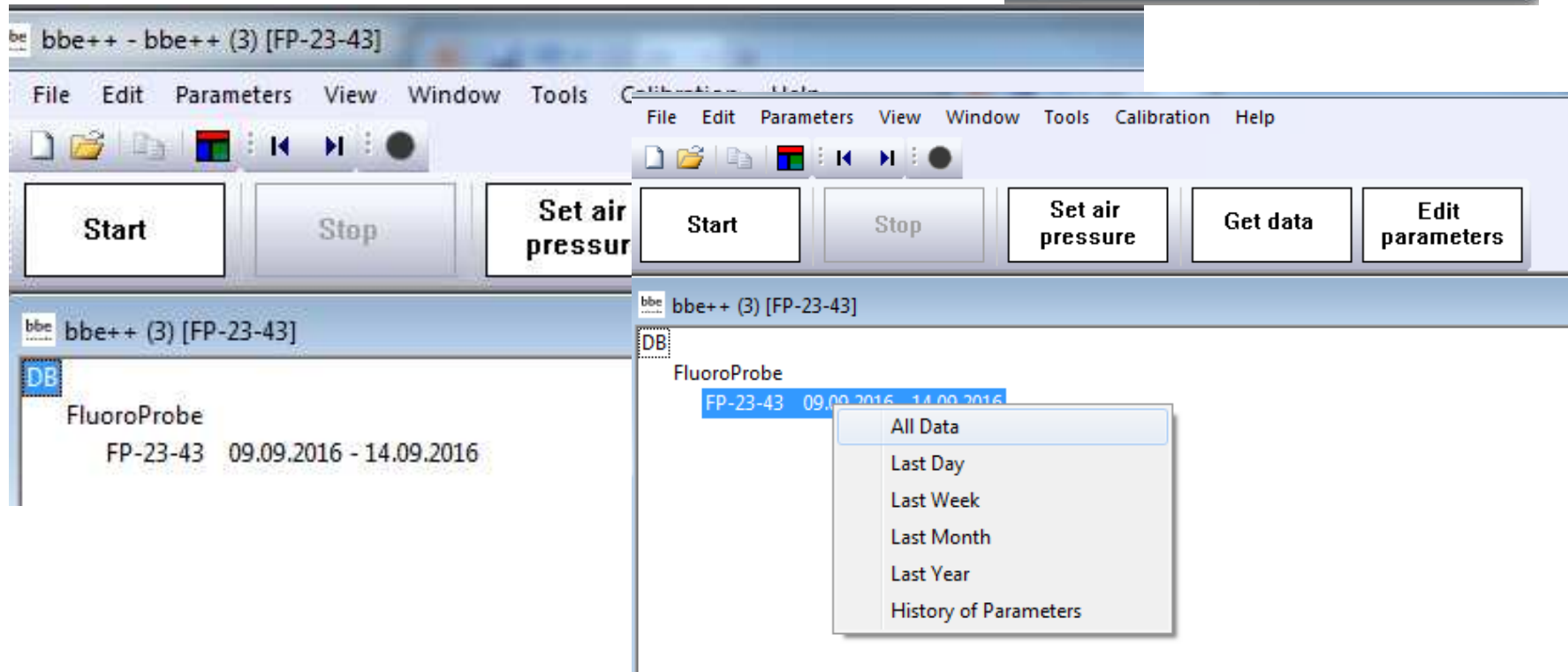
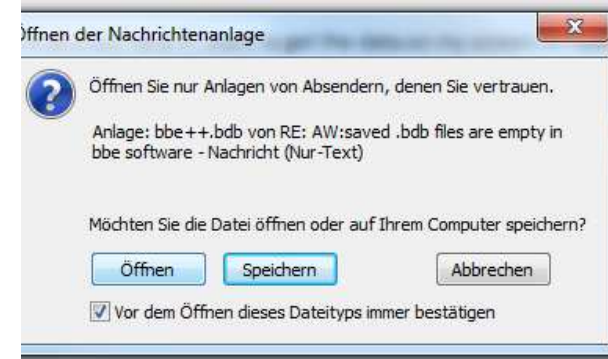
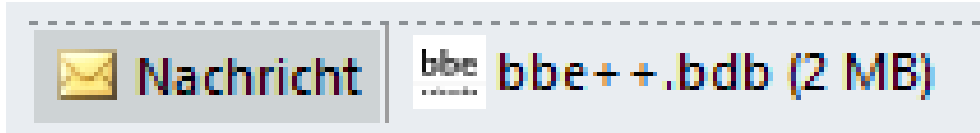
Offsets distilled vs. ultrafiltrated water



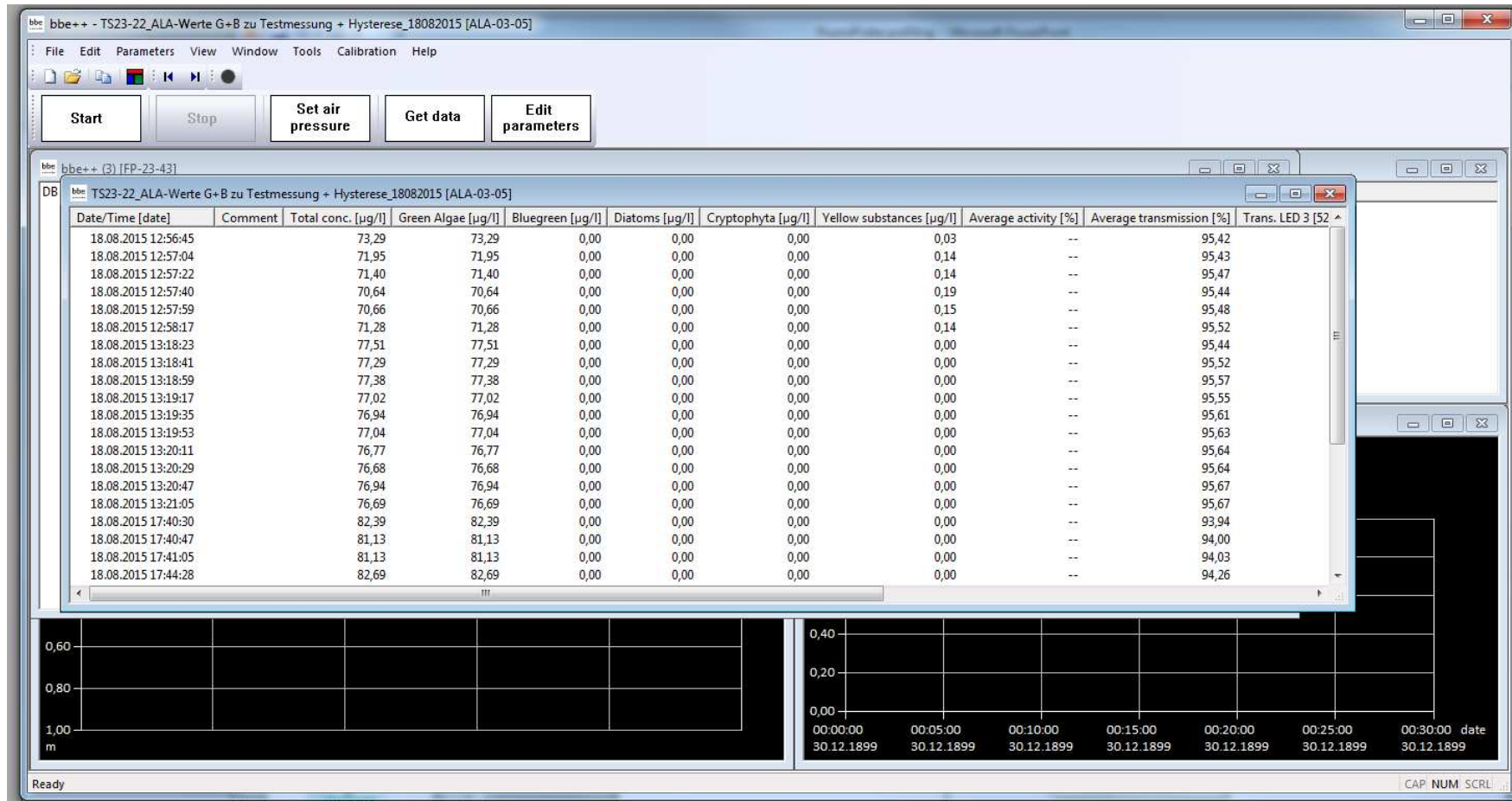
Data transfer



Where to find my data?

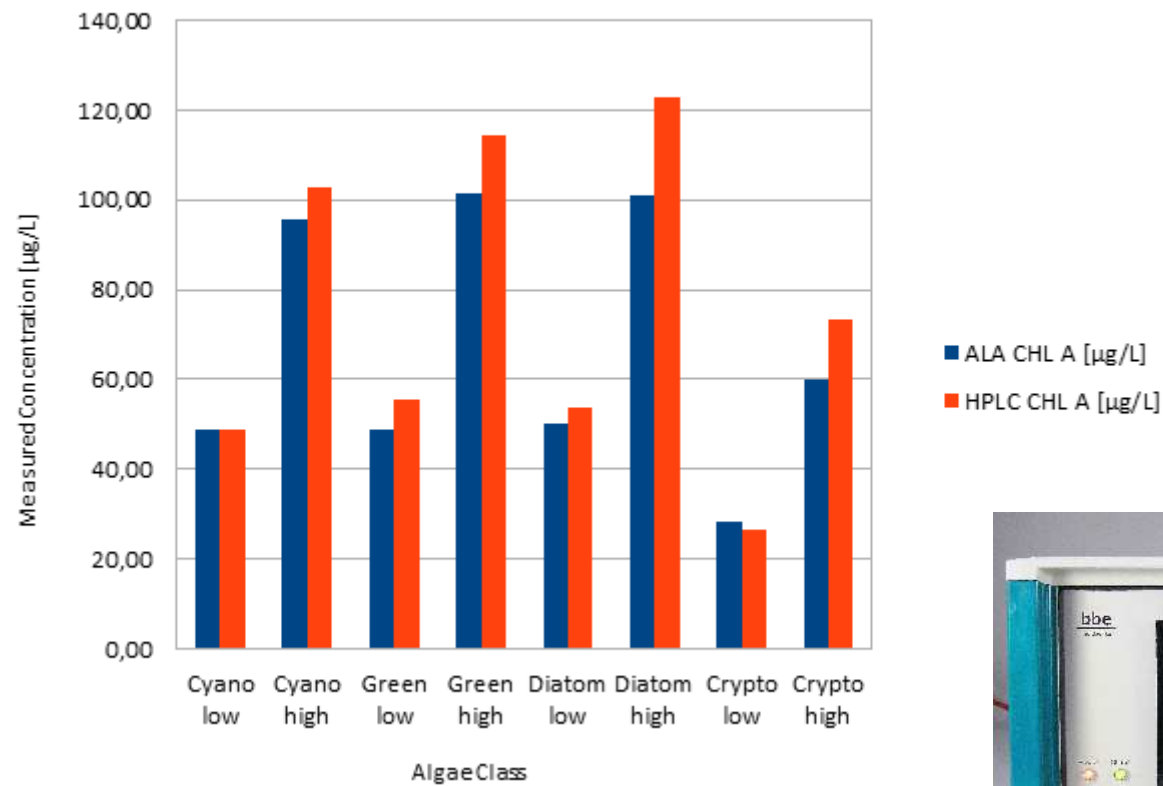


Results



Calibration

Comparison Calibration-ALA with HPLC



Standardized Algae cultures











green algae
bluegreen algae
Diatoms
Cryptophyceae



Chlorella vulgaris
Microcystis aeruginosa
Cyclotella meneghiniana
Cryptomonas sp.

Pigments Important Algae Classes

spectral class	Green algae	Blue-green "algae"	Blue-green "algae" red-type	Green algae	Brown coloured class			Mixed type
	Chlorophyceae Prasinophyceae	Microcystis/ Cyanophyceae	Plankthothrix/ Cyanophyceae	Euglena/ Euglenopyceae	Synura Chrysophyceae	Diatoms Bacillariophyceae	Dinophyceae Dinoflagellates	Cryptophytae/ Cryptomonas
Pigments								
Chlorophyll								
Chlorophyll-a	●	●	●	●	●	●	●	●
Chlorophyll-b	●			●				
Chlorophyll-c					●	●	●	●
Phycobilines								
Phycocyanine		●	●					●
Phycoerythrine		●	●					●
Carotins								
β-Carotin	●	●	●	●	●	●	●	●
Xanthophylls								
Diadinoxanthin				●	●	●	●	
Fucoxanthin					●	●	●	
Lutein	●		●					
Peridinin							●	
Alloxanthin								●
Zeaxanthin	●	●	●	●	●			

FluoroProbe demo-mode in the bbe++ software

