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FluoroProbe III

The Instrument for Depth
Profiles of Microalgae



Rapid and simple
algae monitoring



Suitable for all surface
waters: lakes, reservoirs,
rivers and bathing water



What can the FluoroProbe III do?

The FluoroProbe III is a depth profiler for the rapid detection of chlorophyll-a in phytoplankton



✓ FEATURES

- ▶ Scientific analysis of water quality
- ▶ Qualified measurement of chlorophyll-a
- ▶ Simultaneous measurement of different algal classes
- ▶ Detection of cyanobacteria
- ▶ Depth profiling in real time
- ▶ Evaluation of algal blooms
- ▶ Detection of phytoplankton populations

Water as the basis of life of all organisms has an essential significance. Increasing anthropogenous pollution of water bodies requires an increase in the observation and **assessment of water quality**. Chlorophyll-a is significant in the recognition and evaluation of the characteristic features of a water body since phytoplankton is the main component of biomass present in rivers, lakes and the sea.

The FluoroProbe III measures the chlorophyll-a of suspended phytoplankton *in vivo*. Microalgae of different classes and cyanobacteria (cyanophyceae) are part of a complex ecosystem related to zooplankton and higher organisms. Different algal classes (dinoflagellates) and cyanobacteria are known for their production of biotoxins. Since phytoplankton can be found at various depths in a water body, the estimation of the algal content of the water requires a **rapid and sensitive profile measurement**.

Fluorescence measurements using the FluoroProbe III fulfil these requirements and allow depth profiles to be recorded with the simultaneous analysis of different algal classes, all in real time. The results of the fluorescence spectroscopy of cultured algae and naturally present algae verify the comparability of spectral fluorescence characteristics and serve to evaluate and quantify the chlorophyll-a content of natural populations.

In this way, it is possible to follow the creation and development of algal blooms at high resolutions on the surface as well as at depths. Thus, the FluoroProbe III is the **most suitable instrument for the comprehensive investigation of water bodies** at phytoplankton level and for the determination of potentially harmful cyanobacteria.

How does the FluoroProbe III measure chlorophyll-a content?

Photosynthesis uses the light energy of the sun to gain and convert energy into energy-rich substances. This process is closely linked to the photosystems of algae and cyanobacteria – with chlorophyll-a playing a central role in the conversion. Fluorescence – i.e. the emission of previously absorbed light – is a natural process. When energised by sunlight or artificial light, unused or superfluous energy is immediately given off. The light-collecting photosystem emits red light at a wavelength of 685 - 700nm, which is detected by a highly sensitive photomultiplier. At a suitable excitation the emitted fluorescent light is proportional to the chlorophyll-a content of phytoplankton. In addition to chlorophyll-a, algae and cyanobacteria contain **accessory pigments for the more effective use of light energy** which characteristically influence chlorophyll-a fluorescence.

The use of **six different LEDs** with excitation wavelengths in the visible range of 370 to 610nm improves the calculation and enables a distinction of different algal classes. For a constant excitation over the time of deployment, the FluoroProbe III has an **integrated LED brightness adjustment**. To avoid the influence of ambient light the excitation is **pulsed at high frequency** (6 KHz) so that only the pulsed and filtered fluorescent response is evaluated. The calculation of the distribution of the amount of chlorophyll-a is performed by using the **previously stored fluorescence profiles** (fingerprints) of the known algal classes from an algae library.



MEASUREMENT

- ▶ Chlorophyll-a fluorescence
- ▶ Accessory pigments
- ▶ 6 excitation wavelengths
- ▶ Brightness regulation of excitation
- ▶ High measurement frequency
- ▶ Evaluation based on stored algal spectra



We regularly check our reservoirs using the FluoroProbe. In contrast to lab methods the FluoroProbe delivers a quick overview of algal content in the reservoirs, and the depth profiles in particular help us to decide at what depth we should extract the raw water.



Anita Nienhüser
Drinking Water Department Head
Bergish Water and Environmental
Laboratory of the BTV GmbH

What are the additional features of the FluoroProbe III?

Additional FluoroProbe III data

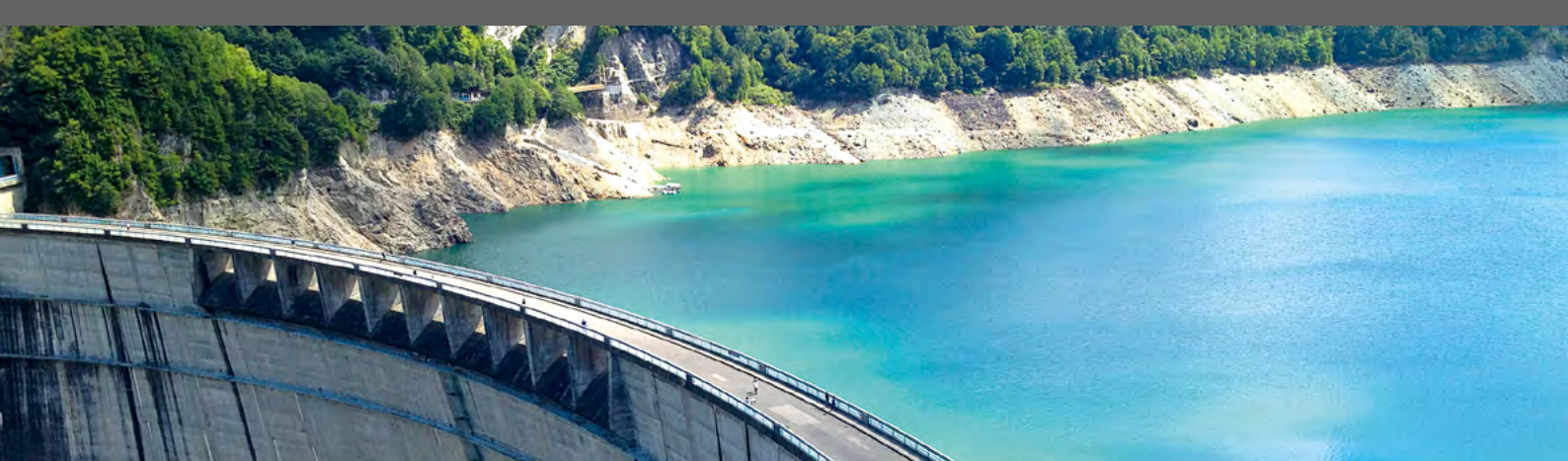
Turbidity-causing particles have an influence on the *in vivo* chlorophyll measurement due to light reduction and reflection. The **optional transmission measurement with automatic turbidity correction** is unique in commercially available chlorophyll measuring instruments and substantially improves the accuracy of the chlorophyll-a measurement. The turbidity values measured are also displayed and stored.

The influence of yellow substances or CDOM, which belong to the group of humic substances, overlaps the fluorescence measurement of chlorophyll particularly at low algal content levels.



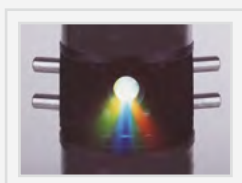
FEATURES

- ▶ Turbidity measurement and compensation
- ▶ CDOM (coloured dissolved organic matter) measurement



FEATURES

- ▶ Calibration using algal cultures
- ▶ Temperature sensor
- ▶ Depth sensor



bbe FluoroProbe III emission lights

This is helped by a **determination of yellow substances and an automatic correction**. The spectral characteristics of yellow substances are stored in the FluoroProbe III and can be **adapted to a specific location** when necessary.

This also applies to algae with particular spectral characteristics: a calibration to a new algal class extends the range of application and increases the accuracy of the allocation of the algal classes. The advantage lies in the use of real algae for the calculation of chlorophyll-a values of natural samples.

For algae and cyanobacteria, nutrients and temperature play a significant role. The ambient temperature in the water is determined by the **optional temperature sensor** on the housing. An exact depth measurement is carried out synchronously using the built-in **pressure sensor**.

Deployment sites of the FluoroProbe III



OUTDOORS

- ▶ Simple operation
- ▶ Settings and analysis with bbe++ software
- ▶ Optionally, handheld for real-time data analysis
- ▶ Submersion depth 0-200 m
- ▶ Huge data memory
- ▶ Cable and cable-free measurements

FluoroProbe III in the field

Using the FluoroProbe III in waterbodies, whether in rivers, reservoirs or lakes, is easy and simple with the **bbe++ software**. The software runs on any current Windows PC and communicates via a USB cable. Optionally, for use of the FPIII **via Bluetooth** a smartphone with corresponding app including a GPS function is available. bbe++ controls all the measurements, the data transfer and the parameterisation. A clear overview allows simple measurement: after measuring the air pressure at the surface (key) for depth measurement, START and STOP signal the beginning and end of a measurement. The FluoroProbe III can be lowered into the water by hand or by using a winch – at a recommended speed of approximately 30 cm/sec. The measurement frequency is up to 4 measurements/second. Maximum depth with cable is 100 m. Real time data are available on the PC or smartphone display. The internal memory contains **10 million datasets**.

The FluoroProbeIII is readily deployable **without cable** by use of an autostart plug. By attaching the plug, the measurement is started and continues until the plug is removed. Via a measurement cable or the USB adapter the data can be transferred to a PC or the USB flash drive supplied. The depth range of the FluoroProbeIII is **down to 200 m**. The internal battery supplies the FluoroProbeIII for extended underwater operation without an external power source. For longer operation bbe recommends the setting for sleep mode with low power usage and an **optional wiper** for the removal of biofilm from the optics. A steel cage has proved useful against mechanical damage during operation from the side of a ship or in rough environments.

Using the FluoroProbeIII in the laboratory

The FluoroProbeIII can also be deployed in the laboratory by using the specially developed Workstation25 as a stand for the instrument. The measurements of chlorophyll-a are performed in a **25ml special cuvette** made of optical glass. The **optional stirrer** motor ensures a homogenous distribution of the sample so that the algae (in particular diatoms) cannot settle. Ambient light is excluded from the optics by a specially designed cover. A test cuvette is also available for function testing.

The FluoroProbeIII can also be deployed in pipe systems using a specially designed **Flow-Through Unit** (inlet and outlet connections). The sample water can be transported using an external pump.

Site of application – what is performed?

The FluoroProbeIII can be used in many different applications: e.g. in the field of **limnology**, lakes and rivers are sampled. Focus is given to ecological questions, in particular changes and status evaluation. Investigations can also be carried out in accordance with the **EU Water Framework Directive**. Moreover, for **quality control** the FluoroProbeIII is also used in reservoirs, particularly for an **analysis of potentially harmful cyanobacteria** *Microcystis aeruginosa* and *Planktothrix rubescens*. In this case, a surface water sample is often insufficient since the cyanobacteria prefer particular depths according to the time of year. The aim is the **prediction of the dynamic of cyanobacteria blooms**. In reservoirs, this concerns the avoidance of cyanotoxins biotoxins. Drinking water producers in Japan, for example, use the FluoroProbeIII integrated in a pipe system. Research trips to the Caribbean and the Antarctic are evidence of the manifold uses of the FluoroProbeIII. Nearshore areas are also used by aquaculture companies in the breeding of seafish. The use of feed and the formation of free nutrients promotes substantial growth of algae with all its undesired consequences. The FluoroProbeIII documents the changes precisely via a high data density.



OUTDOORS

- ▶ Autostart plug
- ▶ Increased depth range
- ▶ Energy control
- ▶ Long-term measurement



ADDITIONAL USES

- ▶ Lab use with the Workstation25 and cuvette
- ▶ Test cuvette for function testing
- ▶ Flow-through measurement



USERS

- ▶ Reservoir operators
- ▶ Aquaculture farms
- ▶ Drinking water producers
- ▶ Assessors
- ▶ Ecologists, Limnologists
- ▶ Oceanographers



The spectral analysis of algae



Chlorophyll-a content from algae and cyanobacteria

- ▶ Reference spectra
- ▶ Fit procedure



Bluetooth-Set with smartphone for data viewing and control



BBE++ SOFTWARE

- ▶ Database
- ▶ Graphic and table representation
- ▶ Macro settings
- ▶ Basic and expert modes
- ▶ Re-calculation
- ▶ Data export

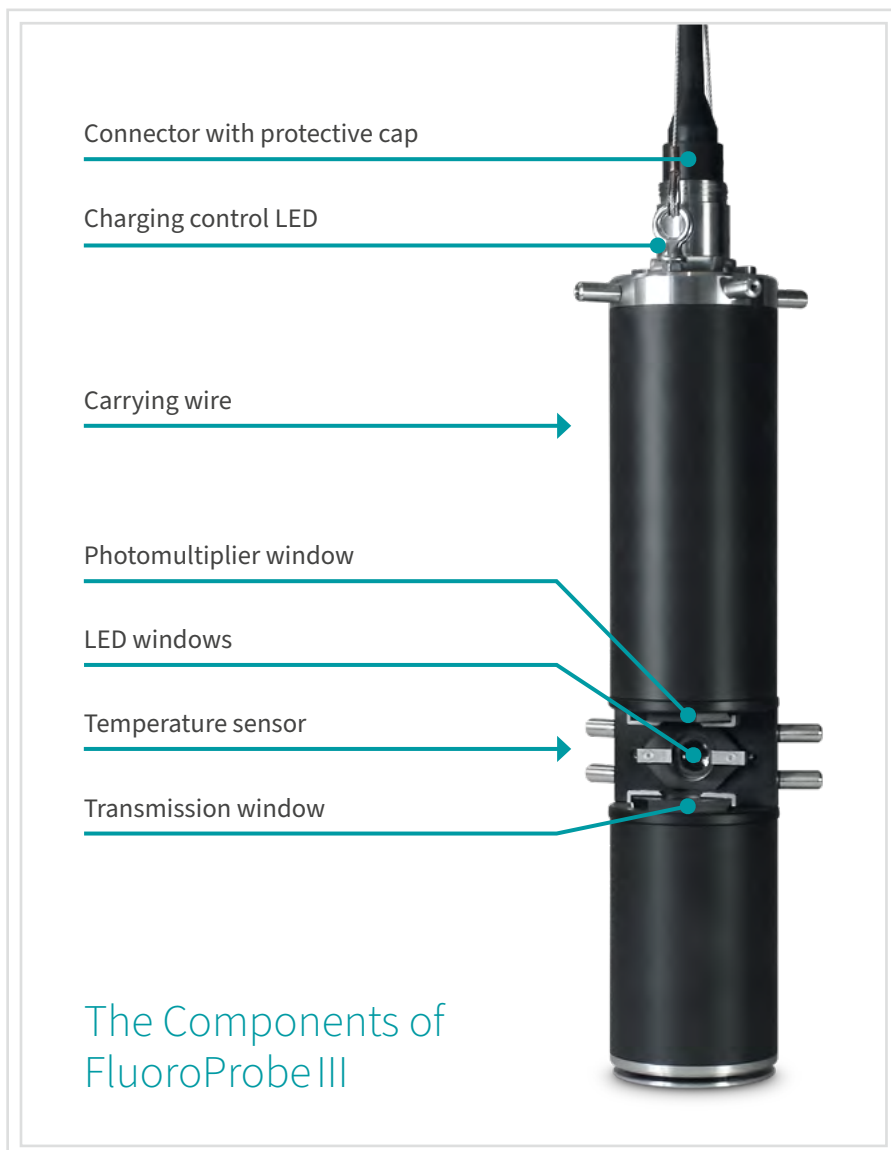
How is the chlorophyll-a calculated?

The FluoroProbeIII is prepared for **immediate chlorophyll determination** by factory calibration using standard algae. This is performed using a special spectrofluorometer (bbe ALA) which has been calibrated using HPLC analyses of extracted algal pigments. HPLC analyses are carried out by external laboratories. These **reference spectra of the algae** used for the determination of the chlorophyll-a are stored in the FluoroProbeIII. Each algal class has a **characteristic fluorescence spectrum** (fingerprint) referring to the chlorophyll content present with regard to form and intensity. Based on the linear combination of the fluorescence signals created by the algal fluorescence of the sample algae, the measurement values obtained are compared with the reference data and **distributed optimally across the different algal classes by means of a fit procedure**. The total chlorophyll-a content results from the sum of the chlorophyll-a of the individual algal classes.

The software allows the user to perform additional calculations by activating or deactivating different algal classes in order to improve the adaptation if necessary. Different reference data from yellow substances can also be applied to the calculation.

The FluoroProbeIII uses the supplied bbe++ software and stores the data in a database. bbe++ takes care of data transmission as well as the **display of the results in graphs and tables**. Macros enable easy pre-selection of suitable display options. The FluoroProbeIII can also be operated and controlled in online mode using the bbe++ software. Access is organised in a clear basic and a complex expert mode with all the possibilities of such advanced software. The basic mode comprises all essential functions for operation in the field and the laboratory. The expert mode allows optimisation of the algal classes via a re-calculation using additional fingerprints. The bbe++ software allows the problem-free **export of data to other programmes** (e.g. Excel).

Components, scope of delivery and maintenance



SCOPE OF DELIVERY

- ▶ The “FluoroProbeIII“
- ▶ Charger
- ▶ USB adapter with USB flash drive
- ▶ Protective cap
- ▶ Light screen
- ▶ Software & Manual
- ▶ RS485/USB cable

Optional:

- ▶ Measuring cables of 10, 20, 30, 50, 100 m
- ▶ Lab cable of 60 cm
- ▶ Cable drum (for 40/100 m cables)
- ▶ Autostart plug
- ▶ Workstation25 with cuvette and stirrer
- ▶ Flow-Through unit
- ▶ Bluetooth set
- ▶ Wiper
- ▶ Steel Cage

Hints on care and calibration

The FluoroProbeIII is **almost completely maintenance-free**. As with all optical instruments the optical path must be kept clean. Rinsing the optics with clean water is sufficient. In case of biofilm, bbe recommends manual cleaning or the use of an optional wiper which cleans the surface periodically between the measurements.

Re-calibration of the FluoroProbeIII is recommended every two years. This can be performed in the calibration laboratory at bbe using standardised algal cultures. A technical inspection which can prevent errors during subsequent field operation can also be carried out at the same time.

For long-term operation at 1 measurement/sec the maximum operation time is 10 hours. If measurements are carried out once per day, the FluoroProbeIII is deployable around 30 days. The maximum charging time is approx. 8 hours.

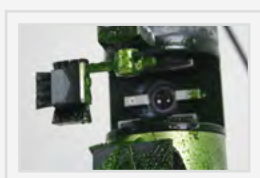


MAINTENANCE & SERVICE

- ▶ Cleaning
- ▶ Calibration, Inspection
- ▶ Operation time
- ▶ Charging

+ ADVANTAGES

- ▶ Replaces a considerable amount of time-intensive laboratory analysis
- ▶ No sample preparation needed
- ▶ Perfect addition to microscopy
- ▶ Real-time analysis of phytoplankton
- ▶ High measuring speed
- ▶ Turbidity correction
- ▶ Yellow substances correction
- ▶ Calibration using algal cultures
- ▶ Use of custom spectra
- ▶ Internal data logger
- ▶ Internal power supply, rechargeable
- ▶ Wide-ranging accessories



FluoroProbe III with Hydro-Wiper after weeks of operation

Technical Details of the FluoroProbe III

| DESCRIPTION | VALUES |
|-----------------------|---|
| Measurands | total chlorophyll [$\mu\text{g chl-a/l}$], green algae [$\mu\text{g chl-a/l}$], cyanobacteria [$\mu\text{g chl-a/l}$], diatoms [$\mu\text{g chl-a/l}$], cryptophyceae [$\mu\text{g chl-a/l}$], yellow substances correction, water temperature (optional), transmission (optional), depth |
| Measuring range | 0 - 500 $\mu\text{g chlorophyll-a/l}$ |
| Measurement principle | spectral fluorometry |
| Resolution | 0.01 $\mu\text{g chlorophyll-a/l}$ |
| Transmission | 0 - 100 % |
| Water temperature | -2 to 40°C |
| Housing material | reinforced plastic / V4A Steel |
| Weight | 6.4 kg (7.2 kg incl. light screen, in water 4.2 kg) |
| Dimensions (H x Ø) | 450 x 140 mm |
| Voltage | 24 V |
| Battery capacity | 3900 mAh |
| Operating time | continuous operation approx. 10 hrs; interval approx. 30 days |
| Memory capacity | 2 GB memory card - 10 million datasets |
| Interface | RS485 and USB |
| Maximum depth | 0 - 100 m (standard) 0 - 200 m (extended) |
| Options | cuvette holder (Workstation25), temperature/transmission measurement, measuring cables: 2 - 100 m, Hydro-Wiper unit, Bluetooth set |

Do you have any questions? Please contact us!

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