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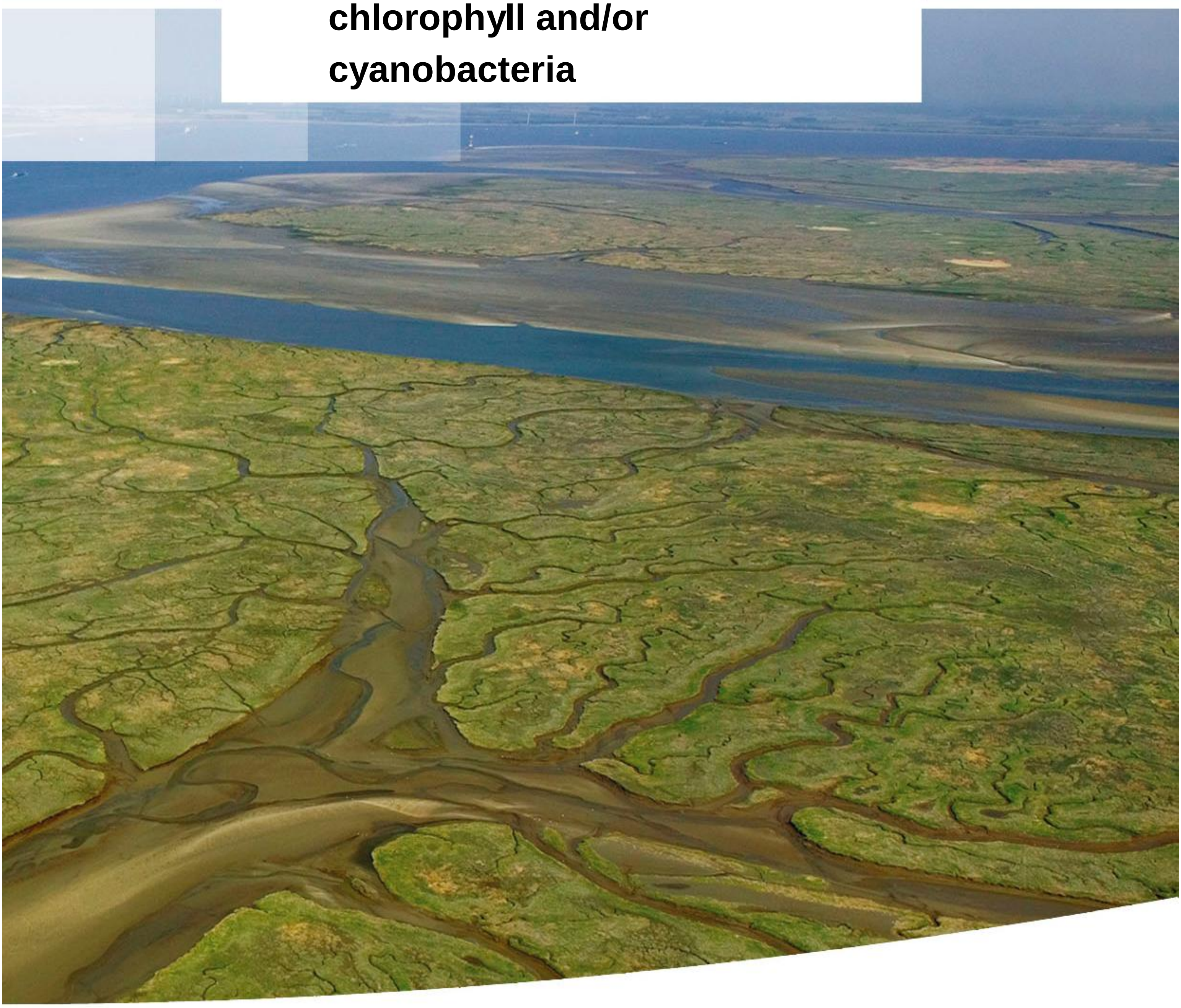
Evaluation of fluorometers for the in situ monitoring of chlorophyll and/or cyanobacteria



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Evaluation of fluorometers for the in situ monitoring of chlorophyll and/or cyanobacteria

Miguel Dionisio Pires

1203593-000

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Client	**Project**	**Reference**
Rijkswaterstaat Waterdienst IJsselmeergebied	1203593-000	1203593-000-ZWS-0004

Key words
Fluorometer, fluorescence excitation, cyanobacteria, chlorophyll, phycocyanin, phycoerythrin

Summary
The quality of surface waters around the world is receiving increased attention. Nevertheless the demand for good quality is increasing and yet in the past surface waters have been badly regulated, leading to severe degradation that may preclude their long-term use. Symptoms of degraded waters could include the occurrence of toxin-producing phytoplankton, like cyanobacteria. Cyanobacteria pose a threat to recreation, drinking water supply and aquatic ecosystems because they produce many different toxins (Chorus & Bartram 1999). Besides toxins, cyanobacteria could produce scums that can reduce sunlight availability.

Monitoring of cyanobacteria (and other phytoplankton) has received much attention over the years. One important monitoring issue has been the need for rapid detection and quantification techniques. In vivo fluorometry is a relatively easy technique that provides quick answers and offers the possibility to collect large quantities of data. It is called in vivo fluorometry because it is based on the direct measurement of the fluorescence of the chlorophyll in the living algal cells. The instruments that measure this fluorescence are called fluorometers.

In the last ten years, many new fluorometers have been introduced on the market. Different fluorometers have different tools, options and prices. Consequently, it is difficult to find the right fluorometer for a specific need. The goal of this study is to make an inventory of several available fluorometers and compare them on reliability, costs and ease of use (for field purposes). For this, 16 fluorometers from nine manufacturers were compared with each other. The question in this study is: what are the advantages and disadvantages of these different fluorometers?

There was a large variation in price between the different fluorometers. The range is from € 1.400,- for the UniLux from Chelsea Technology Groups to € 75.000,- for the LOBO (Satlantic). This large difference mainly has to do with the purpose of the instruments. The cheapest fluorometers are suitable for quick scans and do not monitor other parameters, while the more expensive ones are more suitable for more intense measurements on one or a few locations. The prices of the instruments ascend with the number of possibilities on the instruments.

The difference in weight between the instruments is even larger than the differences in price. The weight is, as for the price, dependent of the purpose of the instrument with the fluorometers with the ones that are meant for long-term monitoring being the heaviest. In the latter case, weight is not a relevant category.

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Only a few instruments come with a complete standard package (i.e. ready to use). These are the instruments from bbe Moldaenke and from Photon Systems Instruments. The standard packages of the Ott/Hydrolab instruments miss many accessories that are needed for the system to be operational.

It is difficult (if not impossible) to pick out one instrument as the best one. For bathing water purposes, the cheap fluorometers (hand-held instruments), like the TriLux and microFlu, are good options. More information (other phytoplankton groups, depth profiling) comes from instruments like the C3, C6, Fluoroprobe, the Ott/Hydrolab instruments and the 6600 V2. For selection of a fluorometer, the purpose of an investigation needs to be defined first. It is also vital that a fluorometer (regardless which one) is frequently calibrated and cleaned.

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Contents

1 Introduction

The quality of surface waters around the world is receiving increasing attention because the demand for good quality is increasing and yet in the past surface waters have been badly regulated, leading to severe degradation that may preclude their long-term use. Symptoms of these degraded waters include the occurrence of toxin-producing phytoplankton, like cyanobacteria. Cyanobacteria pose a threat to recreation, drinking water supply and aquatic ecosystems because they produce many different toxins (Chorus & Bartram 1999).

Monitoring of cyanobacteria (and other phytoplankton) has received much attention over the years. One important monitoring issue has been the need for rapid detection and quantification techniques. For bathing waters where the presence of cyanobacterial blooms is suspected, it is necessary to provide a quick answer. Microscopy, the classical way of quantification, is very labor-intensive (and thus slow reporting) and depends too much on the cell counter (Stowa 2010-18). Other methods (flow cytometry, cell counters, Q-PCR, spectrophotometry) are either also not quick enough, too expensive or still in its development phase.

In vivo fluorometry (IVF) has been by aquatic researchers for several decades. It is called in vivo fluorometry because it is based on the direct measurement of the fluorescence of the chlorophyll in the living algal cells. The same methodology is used to detect the phycobilin pigments of cyanobacteria in water. The benefits of IVF include ease, speed and the ability to collect large quantities of data. There is no special sample handling or processing required, making IVF ideal for real-time data collection. IVF is the easiest method for collecting large quantities of data but there are variables associated with IVF that result in errors and interference. The fluorescence for a given cell concentration is affected by a number of factors including; the amount of light the cell was exposed to prior to the measurement and variation amongst different species, physiological states and environmental conditions. For the most accurate data, IVF data is correlated to quantitative data that can be collected by taking occasional samples to be analyzed for pigment concentration by a technique that is not affected by the conditions of the live sample. Unlike the chlorophylls that have relatively easy and well-established extraction methods (Wright et. al. 1991), phycocyanin and phycoerythrin are water-soluble pigments which makes extractive methods more challenging. Despite these factors, fluorometers are a good monitoring tool because they permit phytoplankton biomass data to be recorded continuously in the field. Fluorometers not only replace the equivalent of hundreds of individual measurements, but fluorometers also have a higher chance of detecting low amounts of phytoplankton. In addition, fluorometers have several advantages over the traditional laboratory analyses. Firstly, with a fluorometer it is possible to obtain data with a high resolution which provides insights in processes with a high turnover rate. Secondly, with a fluorometer non random sampling is avoided. Thirdly, in vivo/in situ measurements do not have the transporation problems of samples. Finally, in situ measurements have the possibility to measure vertical profiles allowing researchers to construct a three dimensional view of the ecosystem.

In the last ten years, many new fluorometers have been introduced on the market. Different fluorometers have different tools, options and prices. Consequently, it is difficult to find the right fluorometer for a specific need. The goal of this study is to make an inventory of several available fluorometers and compare them on reliability, costs and ease of use (for field purposes). For this, 16 fluorometers from nine manufacturers were compared with each other. The question in this study is: what are the advantages and disadvantages of these different fluorometers?

2 The instruments

2.1 Principle of fluorescence excitation

All instruments discussed below make use of fluorescence excitation. This means that when chlorophyll molecules absorb light, a fraction of the energy absorbed is reemitted as fluorescence. Chlorophyll fluorescence techniques are used widely in both laboratory and field studies to assess the abundance and physiological responses of cyanobacteria, microalgae, macroalgae and vascular plants. Chlorophyll a detection supplies data on the total algal biomass (all photosynthetic organisms contain the chlorophyll a pigment). Different types of phytoplankton and cyanobacteria have unique sets of accessory pigments that serve a variety of roles for the organism. These accessory pigments are often unique to a class of algae or cyanobacteria and can be used to identify a specific group. Cyanobacteria contain accessory pigments from the phycobiliprotein family. The primary phycobilin pigments are phycocyanin and phycoerythrin which have strong fluorescent signatures that do not interfere with the fluorescence of chlorophyll. This allows for the *in vivo* detection of cyanobacteria without interference from other groups of algae. Phycocyanin is the predominant phycobilin in freshwater environments while phycoerythrin is the predominant pigment in marine environments. When purchasing an instrument a decision must be made as to which phycobilin pigment the instrument will be configured. Below are some graphical presentations of how fluorometers work for the detection of chlorophyll, phycocyanin and phycoerythrin (Figures 2.1 to 2.3). In general, light of a certain wavelength is emitted and ‘hits’ the phytoplankton cells of the sample. The pigments of these cells absorb the energy of light with a certain efficiency and reemit light (fluoresce) of different wavelengths as absorbed. The energy transfer of a cell (as fluorescence) of a certain wavelength (i.e. contribution of a cell to a specific wavelength; in fluorometers this is in the red area) allows for quantification of different phytoplankton groups (usually cyanobacteria, green algae and diatoms) present in the sample.

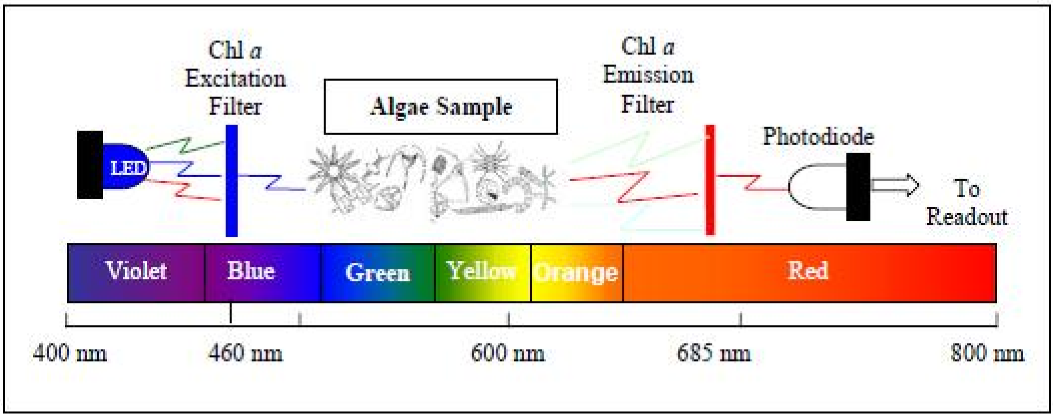

Figure 2.1. Fluorometer configuration for detection of chlorophyll a (Turner Designs 2004)

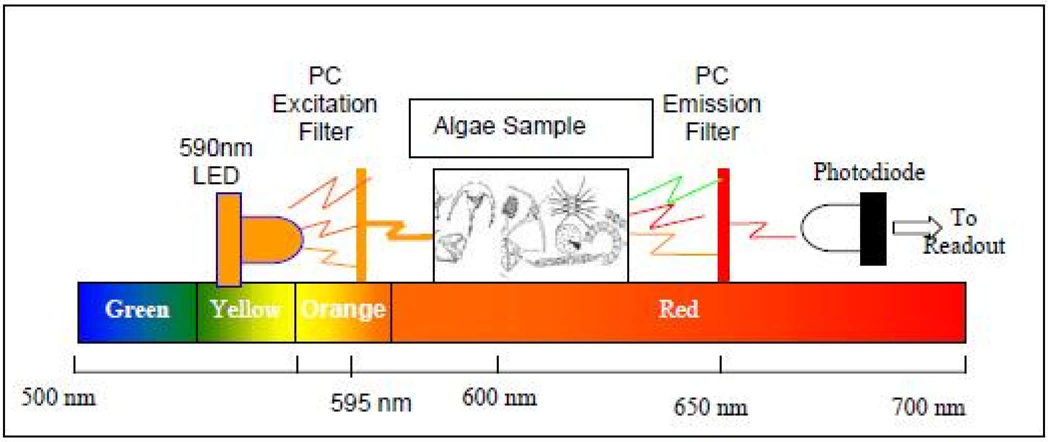

Figure 2.2. Fluorometer configuration for detection of phycocyanin (Turner Designs 2004)

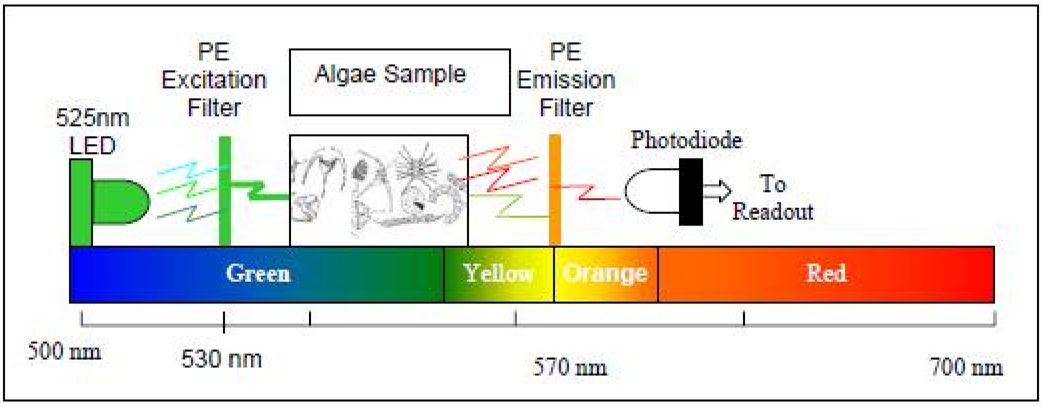

Figure 2.3. Fluorometer configuration for detection of phycoerythrin (Turner Designs 2004)

In the next paragraphs of Chapter 2, different fluorometers will be discussed. They all make use of the principle of fluorescence excitation. A brief description of the different instruments is provided together with their advantages and disadvantages. A separate delivered table (made up in Microsoft® Office Excel) gives an overview of all the here discussed instruments on environment suitability, claims from the manufacturer, costs, ease of use, case studies, and further information. Although all the discussed instruments come with an internal calibration (for chlorophyll, phycocyanin or phycoerythrin), it should be kept in mind that they need to be calibrated occasionally (some more than others). It is also necessary to calibrate fluorometers with phytoplankton species that are representative of the location of interest.

2.2 AlgaeOnlineAnalyzer (bbe Moldaenke)

bbe Moldaenke currently has three fluorometers available: AlgaeOnlineAnalyzer (AOA), Fluoroprobe and AlgaeTorch. The Fluoroprobe and the AlgaeTorch will be discussed in sections 2.2 and 2.3, respectively.

The AOA is capable of measuring four different phytoplankton classes (Chlorophyceae, Cyanophyceae, Cryptophyceae and Bacillariophyceae), due to five Light Emitting Diodes (LED's: excite pigments at 450, 525, 570, 590 and 610 nm wavelengths) and special recorded "fingerprints" for each algae class. The measuring cell is automatically and periodically cleaned using a motorized piston. The AOA takes humic substances into account (which can interfere with the fluorescence from the Bacillariophyceae) with a specific UV LED.

Advantages
Two studies (Izydorczyk et al. 2009, Cagnard et al. 2006) show good correlation of the AOA with conventional methods (cell counts, spectrophotometry). For the determination of cyanobacteria, no additional equipment (like a sensor) is needed.

Disadvantages
The AOA needs a steady platform for operation. The AOA is more suitable as an online laboratory fluorometer.

2.3 FluoroProbe (bbe Moldaenke)

The operation procedure of the Fluoroprobe is the same as that for the AOA. The Fluoroprobe can therefore be seen as the field equivalent of the AOA.

Advantages
Realistic estimations of algae *in situ* (after appropriate calibration, Leboulanger et al. 2002) and the results are very robust (Stowa 2010-18). The Fluoroprobe is also available as a laboratory fluorometer. For the determination of cyanobacteria, no additional equipment (like a sensor) is needed.

Disadvantages
Its weight (4.6 kg) makes it less easy to perform analyses in the field than other fluorometers (like the AlgaeTorch which is also from bbe Moldaenke).

2.4 AlgaeTorch (bbe Moldaenke)

The AlgaeTorch can be seen as the “little brother” of the Fluoroprobe. This is not only because it is lighter than the Fluoroprobe but also because it does not measure five different phytoplankton classes but only two (cyanobacteria and other microalgae) because it only has three LED’s instead of five like the Fluoroprobe (and the AOA). The AlgaeTorch is explicitly designed for the measurement of cyanobacteria. AlgaeTorch and Fluoroprobe give similar results when measuring cyanobacteria from field samples (Newsletter of the Cyanobacteria Workgroup No. 11, May 2010).

Advantages
Due to its weight of 1.2 kg, it is easier to handle in the field than the Fluoroprobe. Results are stored in the internal memory for later analyses at the lab. No external battery is needed. The Torch itself can be charged. For the determination of cyanobacteria, no additional equipment (like a sensor) is needed.

Disadvantages
Does not give a warning when the measurements (of for instance cyanobacteria) are too high. If interested in other phytoplankton species, than this apparatus is not suitable.

2.5 YSI 6600 V2

YSI provides multiparameter water quality sondes for real-time monitoring. For this study the 6600 V2 sondes were chosen because they can monitor chlorophyll, phycocyanin (for measurements of cyanobacteria in freshwater systems) or phycoerythrin (for measurements of cyanobacteria and red algae in marine and estuarine systems). Measurements of chlorophyll, phycocyanin or phycoerythrin are not part of the standard measuring parameters. For this, it is necessary to use specific sensors for all three pigments: the 6025 sensor for chlorophyll, 6131 for phycocyanin and 6132 for phycoerythrin.

Advantages
The 6600 V2 has a wiped sensor technology to reduce biological fouling. Solid standard available to check sensor stability.

Disadvantages
The V2 sonde is heavy (> 3 kg). Sensors for chlorophyll and cyanobacterial pigments need to be installed.

2.6 YSI 6600 EDS V2

The 6600 EDS V2 operates in the same way as the 6600 V2, but was developed for severe fouling environments. Sensors for chlorophyll, phycocyanin and phycoerythrin are the same as for the 6600 V2. There are no case studies known that have been published in peer-reviewed scientific journals. On the website of YSI different cases ("application notes") are published in which the V2 sondes (V2 and EDS V2) in combination with the chlorophyll and/or phycocyanin/phycoerythrin sensors have been successfully applied (like a vertical profiling study for the safety of drinking water by Dr. Burkholder from North Carolina State University.

Advantages
Like 6600 V2, the 6600 EDS V2 version has a wiped sensor technology for minimizing fouling Solid standard available to check sensor stability.

Disadvantages
The sonde is heavy (> 3 kg). Different sensors for chlorophyll a and cyanobacterial pigments need to be installed.

2.7 microFLU (TriOS Optical Sensors)

The microFlu fluorometers are low-cost miniaturized submersible fluorometers. Three different sensors are available: for CDOM (microFlu-CDOM), chlorophyll (microFlu-chl) and cyanobacteria (microFlu-blue). They are very sensitive (cyanobacteria for example are measured with a 0.02 µg/L precisement) and very light (between 0.5 -0.7 kg, depending on the housing). TriOS provides three different configurations for the microFlu-blue and microFlu-chl sensors: a configuration set for use in laboratories (costs € 4.400,-), a configuration set in which the apparatus remains permanently outside (costs € 8.000,-) and a configuration set for field measurements (costs € 6.500,-). The sensor itself (either chlorophyll or phycocyanin) costs about € 3.300,-. The difference between the last two sets is that the first one uses an attached transmitter with a touch screen while the last one is every time carried into the field. This last one sends data trough bluetooth to a PDA or smart phone. Installation software for the PDA or smart phone is provided. Data can be stored with gps coordinates. It is possible to measure chlorophyll and phycocyanin on one fluorometer at the same time for the laboratory and the permanently outside setup using a TriBox. This is not an option for the field measurements setup.

Advantages
The microFlu-blue is very sensitive to variations of cyanobacteria quantity (Cagnard et al. 2006). Low power consumption. Special coating for fouling protection. Lightweight (0.5-0.7 kg). They seem to be easy to handle (straightforward measuring).

Disadvantages
Needs, at least, monthly calibration (Cagnard et al. 2006). A PDA, smart phone or laptop/PC is needed. The field measurements configuration cannot simultaneously measure chlorophyll and phycocyanin.

2.8 Multi-Exciter (JFE Advantech Co.)

The Multi-Exciter is manufactured by the Japanese firm JFE Advantech Co. As other equipments here described, the Multi-Exciter emits modulated lights of different wavelengths for the discrimination of phytoplankton groups (cyanophyceae, bacillariophyceae and chlorophyceae, which by JFE are incorrectly termed as different algal species). There is almost no information available on the Multi-Exciter. The few things mentioned are the weight (760 g), a depth range of 200 m, excitation wavelengths (375, 400, 430, 470, 505, 525, 568 and 590 nm). The Multi-Exciter uses more excitation wavelengths than other fluorometers and should (in theory) be able to distinguish more phytoplankton groups and provide a sharper distinction between them. Costs are unknown (JFE did not respond to questions on this). From the website, it looks that the Multi-Exciter needs to be plugged into a laptop. No case studies are known.

Advantages
Several wavelengths that may lead to sharper distinctions between phytoplankton groups. Lightweight.

Disadvantages
Not enough information provided by the manufacturer, which is not a good sign. Needs a laptop (or PC) for operation.

2.9 Hydrolab Multiparameter MS5 probe (Ott)

Ott provides the Series 5 Multiparameter probes (MS5, DS5 and DS5X) which were designed by Hydrolab (nowadays Hach Hydromet). The DS5 and DS5X will be discussed in paragraph 2.9. Here we focus on the MS5, the smallest probe. The MS5 probe is capable of measuring a number of parameters simultaneously. The sensors are not part of the standard platform and should be selected at the time of ordering. The sensor for cyanobacteria measures both phycocyanin (excitation at 590 nm, emission at 650 nm) and phycoerythrin (excitation at 530 nm, emission at 570 nm). The costs of the MS5 probe with three sensors (including chlorophyll and cyanobacteria sensors) is between € 13.000,- and € 15.000,-. The standard equipment is the probe (without sensors) and electronics. Important options are an underwater cable and internal memory. In case it is impossible to directly connect it to a laptop or data-logger a Windows handheld can be acquired. Not many case studies with cyanobacteria are known. A MS5 system was used by Deltares to study Vlietland Lake and measure the effectiveness of cyanobacterial growth reduction methods (personal communications R.E. Trouwborst, 2010).

Advantages
The possibility to measure several parameters. Lightweight (1.3 kg). Compact instrument. Also suitable for groundwater measurements. Solid standard available to check sensor stability.

Disadvantages
Almost no standard equipment. If the purpose is to measure under water then a special cable needs to be bought.

2.10 Hydrolab Multiparameter DS5 and DS5X probe (Ott)

The DS5 and DS5X probes function in the same as the MS5. Both probes can measure up to 15 parameters simultaneously. The DS5 is suitable from point-to-point measurements while the DS5X is suitable for long-term use for which it has a brush motor to clean the sensors before measurements, so that maintenance is minimized. The sensors for DS5 and DS5X are the same as for the MS5. Costs of the DS5 and DS5X with three sensors (as done for the MS5 above) are € 14.000,- to € 16.000,- and around € 17.000,-, respectively. No case studies with cyanobacteria are known.

Advantages
The possibility to measure several parameters. Compact instrument. Also suitable for groundwater measurements. DS5X has wiped sensor technology to reduce biological fouling. Copper coated sensors available to reduce biological sensor fouling

Disadvantages
Almost no standard equipment. If the purpose is to measure under water then a special cable needs to be bought. DS5 and DS5X are heavier then the MS5 (3.4 kg).

2.11 Algal Online Monitor (Photon Systems Instruments)

The Algal Online Monitor (AOM) is a portable and robust device for online detection and continuous monitoring of photosynthetic microorganisms in both natural and artificial water bodies. Photon Systems Instruments claims that it detects and discriminates among cyanobacteria, green and brown algae, diatoms, and other microbes. Case studies however were not provided. AOM is very sensitive (30 ng Chl/L) which allows early detection of very low concentrations of photosynthetic organisms. Although the manufacturer states that the AOM is portable, the equipment requires to be fixed on a wall or a table. This makes it unsuitable for point-to-point measurements. It is unclear if a long cable can be used for depth profiles. The costs are € 12.990,-.

Advantages
Very low detection limit (30 ng Chl/L).

Disadvantages
Needs fixing on a wall or table. Almost no information available to make a good evaluation. Not very light (3.4 kg).

2.12 LOBO (Wetlabs/Satlantic)

LOBO (Land/Ocean Biogeochemical Observatory) is an integrated real-time, water quality monitoring package developed at the Monterey Bay Aquarium Research Institute. LOBO houses a suite of *in situ* sensors to measure water properties (both in marine and freshwater systems) including physical (e.g. temperature, depth, salinity and turbidity), chemical (CDOM, nitrate and dissolved oxygen) and biological parameters (chlorophyll fluorescence and optionally ancillary pigments). The LOBO needs a floating platform (is part of the standard package) and it comes complete with a wireless telemetry system, integrated sensor suite, automated processing and archiving software. The sensors have an anti-biofouling system. The case studies shown on the website of Satlantic show the floating system in the middle of rivers, bays or other aquatic systems where the LOBO's have been monitoring continuously for a few years. A buoyant/floating system with solar power is also available from YSI and Hydrolab/Ott including the possibility for online data transport.

Advantages
Floating system. Uses solar radiation for power. Once the software is installed and the system is floating data are immediately live on the web.

Disadvantages
The LOBO is expensive (about € 75.000,- including a phycocyanin sensor which is not part of the standard equipment) and heavy (around 70 kg). The system is not suitable for a quick measurement and to travel from one system to another. It is designed for long-term monitoring on one location.

2.13 C3 Submersible fluorometer (Turner Designs)

The C3 Submersible Fluorometer is designed to incorporate up to three optical sensors ranging from the ultraviolet to the infrared spectrum, but it is also possible to choose for a fluorometer with one or two optical options. Internal memory storage capacity combined with an external submersible lithium ion battery allows the C3 to run during extended or short-term deployments. Each C3 comes with a factory-installed temperature sensor and a depth rating of 600 meters. Optional factory-installed depth sensors and mechanical wiper are also available. The housing is highly resistant to harsh environments. The costs for C3 with three optical sensors is around € 9.500,- which includes (as an option) a mechanical wiper, battery pack and a 5 m cable. An extra 5 m cable is about € 500,-. 25 and 50 m cables are more expensive. An interesting option on the C3 is the C-ray deployment body that enables horizontal towing (from for instance a boat) allowing spatial measurements. A case study on Turner's website showed its ability to detect changes in algal abundance, track an algal bloom, and characterize the water column.

Advantages
Robust system. Possibility for installation of up to three optical sensors for a good price. Horizontal towing possible. Internal storage already supplied. Not heavy (1.64 kg). Low detection limit.

Disadvantages
Cables and battery pack are an option.

2.14 C6 platform for Cyclops-7 sensors (Turner Designs)

The C6 Multi-Sensor Platform integrates up to six Cyclops-7 fluorescence and turbidity sensors for extended or short-term deployments. The C6 provides individual automatic gain control, calibration, digital data reporting and data logging for each Cyclops-7 sensor. Each C6 comes with factory-installed temperature, pressure sensors and a depth rating of 600 meters. The costs are around € 10.500,- for a platform with a mechanical wiper, 3 sensors (for chlorophyll, phycocyanin and phycoerythrin), battery pack and 5 m cable (all options, including the sensors). As for the C3 it is possible to get longer cables (same prices). A published case study on the website of Turner shows a positive report and mentions incorporation of the Cyclops in future studies (of the Southampton Oceanography Centre, UK).

Advantages
Robust system. Internal storage already supplied. The Cyclops sensors have a low detection limit.

Disadvantages
Heavier than the C3 (2.74 kg). Cables, sensors and battery pack are an option.

2.15 UniLux and TriLux (Chelsea Technology Groups)

The UniLux is a single parameter fluorometer which can detect one of chlorophyll-a, phycocyanin or phycoerythrin. It is necessary to connect the UniLux to a PC, laptop or a PDA (using a Roamer Wireless Data-Link) to operate it.
The TriLux operates in a different fashion to the single wavelength fluorometer (UniLux), providing information about the proportion of different light harvesting pigments in the algal sample being interrogated. The TriLux operates on the principle that energy absorbed by the light harvesting pigments is rapidly transferred to chlorophyll-*a* (like other multi-wave fluorometers (e.g. AOA) here discussed). A proportion of the energy absorbed, however, is re-emitted as chlorophyll-*a* fluorescence with a peak of 685nm. All versions of TriLux come with a chlorophyll-*a* channel as standard, two other channels can then be selected between turbidity, phycoerythrin, and phycocyanin. The TriLux allows the user to assess the relative contribution to Chlorophyll-*a* fluorescence emission from the different light harvesting pigments absorbing light at each of the chosen excitation wavelengths. This information can then be used to assess the different classes of phytoplankton present in the sample. Phycoerythrin and phycocyanin data from TriLux is related to the Chlorophyll-a fluorescence emission. Users who require phycoerythrin and phycocyanin concentration data may require the individual CTG UniLux fluorometers.

Advantages
UniLux and TriLux have a mechanical wiper system to keep them clean of deposits and bio-fouling. Lightweights (100 g). Relatively cheap equipment (around € 1.400,- for the UniLux and € 1.700,- for the TriLux).

Disadvantages
The UniLux and TriLux need to be connected to a laptop or PC to be able to operate them. In addition, a cable is needed to connect UniLux and TriLux to the laptop or PC, which is not part of the standard package. The UniLux can only measure one parameter (chlorophyll or phycocyanin for example). For simultaneous measurements of chlorophyll, phycocyanin and phycoerythrin a TriLux is needed.

3 Results and discussion

The description of the compared instruments shows that all of them have certain advantages and disadvantages. Some are more suitable for field work, especially for point-to-point measurements than others are. The instruments also differ in price (range: € 1.400,- tot € 75.000,-, not including the Multi-Exciter) and weight (from 100 g to 70 kg). Finally, there are also differences in the completeness of the instruments, i.e. how many options need to be bought to have an operational instrument. In this chapter, the instruments were ranked according to price, weight and completeness. Completeness of instruments was given priority over price and weight (and price in its turn over weight).

There is a large variation in price between the different fluorometers. The range is from € 1.400,- for the UniLux from Chelsea Technology Groups to € 75.000,- for the LOBO (Satlantic) (Table 3.1, in which only no price information was obtained for the Multi-Exciter). This large difference mainly has to do with the purpose of the instruments. The UniLux for instance, is suitable for a quick tour from one location to another. However, it is (probably) not suitable for depth profiles and it is not possible to monitor other parameters as well. The LOBO on the other end is not suitable for monitoring at different locations within one or a few days. It is designed to be placed in the water at one location and to depth profiles for longer periods for several parameters. The prices of the instruments ascend with the number of possibilities on the instruments.

Table 3.1. Prices of different fluorometers and ranking (lowest is best).

Instruments	Manufacturer	Price	Rank
UniLux	Chelsea	€ 1.400,-	1
TriLux	Chelsea	€ 1.700,-	2
microFlu	TriOS	€ 6.500,-	3
AlgaeTorch	bbe Moldaenke	€ 7.500,-	4
C3	Turner	€ 9.500,-	5
6600 EDS V2	YSI	€ 10.000,-	6
6600 V2	YSI	€ 10.500,-	7
C6	Turner	€ 10.500,-	7
AOM	Photon Systems Instruments	€ 12.990,-	8
MS5	Ott	€ 14.500,-	9
DS5	Ott	€ 15.500,-	10
DS5X	Ott	€ 17.000,-	11
Fluoroprobe II	bbe Moldaenke	€ 23.000,-	12
AOA	bbe Moldaenke	€ 27.500,-	13
LOBO	Satlantic	€ 75.000,-	14
Multi-Exciter	JFE	?	15

The purchase costs are only relevant for the hand-held instruments (e.g. UniLux and microFlu) and not for the systems on a platform/probe (e.g. LOBO, the YSI and Ott instruments). For instruments on a platform, the operational costs are important because these instruments require frequent calibration (as for the hand-held instruments) and cleaning. The platform instruments remain for long periods in the water and accumulate much biofouling in time which may interfere with their measuring accuracy. EGV measurements from Delfland Water Board show that after two weeks deviations are observed due to fouling.

These cleaning costs exceed the purchase costs of the instrument (personal communications R.E. Trouwborst, 2010).

The difference in weight between the instruments is even larger than the difference in prices (Table 3.2). The least expensive instruments more or less are the lightest. The weight is, as for the price, dependent of the purpose of the instrument with the fluorometers with the ones that are meant for long-term monitoring being the heaviest. Naturally, the fluorometers that are best suited for short term monitoring at several locations are the lightest ones. Weight is only relevant however, for the hand-held instruments and not for systems like the LOBO and AOM. The high ranking of these instruments should therefore be interpreted with care.

Table 3.2. Weights of fluorometers and ranking (lowest is best). The asterisk denotes an estimate.

Instruments	Manufacturer	Weight (kg)	Rank
UniLux	Chelsea	0.1	1
TriLux	Chelsea	0.1	1
microFlu	TriOS	0.5	2
AlgaeTorch	bbe Moldaenke	1.2	4
C3	Turner	1.64	6
6600 EDS V2	YSI	3.18	8
6600 V2	YSI	3.18	8
C6	Turner	2.74	7
AOM	Photon Systems Instruments	3.4	9
MS5	Ott	1.3	5
DS5	Ott	3.4	9
DS5X	Ott	3.4	9
Fluoroprobe II	bbe Moldaenke	4.6	10
AOA	bbe Moldaenke	19	11
LOBO	Satlantic	70	12
Multi-Exciter	JFE	1*	3

An important aspect for the choice of a fluorometer is if the standard package is complete or if accessories still need to be purchased. In Table 3.3, the different instruments were ranked for completeness of the standard package (yes/no). For the instruments that were not complete, the missing accessories were ranked for importance: tools for data reading and analyzing (like a data-logger or a laptop) were ranked as the most important missing tool, followed by cables (for connection to data reader and for depth profiles) and other accessories. The complete instruments were ranked as best of course. These are: AlgaeTorch, Fluoroprobe, AOA (all bbe Moldaenke), 6600 V2, 6600 EDS V2 (both from YSI), AOM (Photon Systems Instruments) and LOBO (Satlantic). Once purchased, these systems are ready to measure. The instruments for which the standard package was incomplete, the C3 from Turner scored best. The C3 misses cables for depth profiles and a battery pack needs to be bought before it can operate. This battery pack costs around € 2.000,-. The instruments of Ott (MS5, DS5 and DS5X) scored least because at purchasing one of the Ott instruments, only the sonde (+ electronics) and (chosen) sensors are delivered. Batteries, an underwater cable, internal memory and a data reader are accessories.

Table 3.3. Completeness at acquisition of fluorometers and ranking (lowest is best). Fluorometers were ranked according to the accessories that need to be bought to make them operational. Ranking in importance of accessories was given as follows (from most severe to least): data reader (laptop or data logger), cables and other. For missing accessories, see Chapter 2.

Instruments	Manufacturer	Complete?	Rank
UniLux	Chelsea	No	3
TriLux	Chelsea	No	3
microFlu	TriOS	No	3
AlgaeTorch	bbe Moldaenke	Yes	1
C3	Turner	No	2
6600 EDS V2	YSI	Yes	1
6600 V2	YSI	Yes	1
C6	Turner	No	4
AOM	Photon Systems Instruments	Yes	1
MS5	Ott	No	5
DS5	Ott	No	5
DS5X	Ott	No	5
Fluoroprobe II	bbe Moldaenke	Yes	1
AOA	bbe Moldaenke	Yes	1
LOBO	Satlantic	Yes	1
Multi-Exciter	JFE	no	3

The different rankings from Tables 3.1 to 3.3 were summed up in Table 3.4. The lower the sum of ranks is the better the overall score is of an instrument. Best score is for the UniLux (sum of ranks: 5). Low scores can also be seen for TriLux (also from Chelsea), microFlu (TriOS) and AlgaeTorch (bbe Moldaenke). All of them are hand-held instruments. Does this mean that these instruments are the best? It still depends on the goal or purpose of the investigation for which a fluorometer will be purchased. These four instruments with a low ranking also have their limitation (see also Chapter 2). UniLux and AlgaeTorch only monitor cyanobacteria and not other phytoplankton. Working with the microFlu needs sensor switching if one wants to measure both chlorophyll and phycocyanin. The TriLux is the only instruments of these four that can measure chlorophyll, phycocyanin and phycoerythrin at the same time. Disadvantage however, is that depth profiles are not possible (only AlgaeTorch is suitable for this when comparing these four fluorometers).

Table 3.4 also does not suggest that instruments with a high sum of ranks (like the LOBO and the AOA) are less capable equipments. Their design is simply meant for other type of studies than for instance a quick scan for bathing water quality, for which instruments like UniLux and microFlu may be suitable. An instrument like the LOBO for example is designed for long-term studies with depth profile measurements and involvement of several parameters. Others, like the AOM from Photon Systems Instruments are better designed for drinking water purposes. The choice of a fluorometer therefore depends on the type of study.

Table 3.4. Sum of ranking of Tables 3.1 to 3.3.

Instrument	Manufacturer	Ranking (price)	Ranking (weight)	Ranking (completeness)	Sum of ranks (lowest is best)
UniLux	Chelsea	1	1	3	5
TriLux	Chelsea	2	1	3	6
microFlu	TriOS	3	2	3	8
AlgaeTorch	bbe Moldaenke	4	4	1	9
C3	Turner	5	6	2	13
6600 EDS V2	YSI	6	8	1	15
6600 V2	YSI	7	8	1	16
C6	Turner	7	7	5	19
AOM	Photon Systems Instruments	8	9	1	18
MS5	Ott	9	5	6	20
DS5	Ott	10	9	6	25
DS5X	Ott	11	9	6	26
Fluoroprobe II	bbe Moldaenke	12	10	1	24
AOA	bbe Moldaenke	13	11	1	25
LOBO	Satlantic	14	12	1	27
Multi-Exciter	JFE	15	3	3	21

Besides price, weight and completeness other categories may be used for the comparison of fluorometers. These other categories were not taken up in this study due to the fact that this was a quickscan of fluorometers. However, one may want to compare the different instruments for different reasons using the following categories: hand-held vs measure station, detection limit, sensor quality and stability, communication possibilities, depth range, fouling, software provided and manual (e.g. user friendliness). Hand-held instruments, like Uniflux or microFlu, would not score well as a measure station. Also for communication possibilities, the more elaborate systems like the LOBO will score better than the cheapier (hand-held) instruments. As stated above, the choice of a fluorometer therefore depends on the type of study.

4 Conclusions and recommendations

The goal of this study was to make an inventory of several available fluorometers. For this, a quick scan on advantages and disadvantages was made of 16 fluorometers that are on the market.

It is difficult (if not impossible) to pick out one instrument as the best one. As discussed above, the choice of a fluorometer depends on the purpose of the investigation. However, in this study the purpose was to compare the instruments for easiness in the field. In that case, the AOA from bbe Moldaenke and the AOM from Photon Systems Instruments were classified as not very suitable.

When considering the other instruments then the cheaper instruments, like TriLux and microFlu, are good options for investigation of algae in relation to bathing water quality. If one wants more information (on different phytoplankton groups and depth profiles) then instruments like the C3, C6, Fluoroprobe, the Ott instruments and the 6600 V2 are more appropriate. Finally, if the intention is to have information on a water body for an extended period (up to several years) for a large parameter set (for instance to get chlorophyll or phycocyanin measurements integrated with physical and chemical data) but not traveling to the location every day, then the LOBO would be the most appropriate.

The first recommendation therefore is to define the purpose of the investigation and then select a fluorometer. Chapter 2 in combination with Tables 3.1-3.3 may assist in the choice of a fluorometer.

It is necessary that a purchased fluorometer is frequently calibrated by discrete sample collection and with species representative of the location of interest.

5 References (including studies mentioned in separately delivered table)

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