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# Quantitative filter technique measurements of spectral light absorption by aquatic particles using a portable integrating cavity absorption meter (QFT-ICAM)

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## Abstract:

The accurate determination of light absorption coefficients of particles in water, especially in very oligotrophic oceanic areas, is still a challenging task. Concentrating aquatic particles on a glass fiber filter and using the Quantitative Filter Technique (QFT) is a common practice. Its routine application is limited by the necessary use of high performance spectrophotometers, distinct problems induced by the strong scattering of the filters and artifacts induced by freezing and storing samples. Measurements of the sample inside a large integrating sphere reduce scattering effects and direct field measurements avoid artifacts due to sample preservation. A small, portable, Integrating Cavity Absorption Meter setup (QFT-ICAM) is presented, that allows rapid measurements of a sample filter. The measurement technique takes into account artifacts due to chlorophyll-*a* fluorescence. The QFT-ICAM is shown to be highly comparable to similar measurements in laboratory spectrophotometers, in terms of accuracy, precision, and path length amplification effects. No spectral artifacts were observed when compared to measurement of samples in suspension, whereas freezing and storing of sample filters induced small losses of water-soluble pigments (probably phycoerythrins). Remaining problems in determining the particulate absorption coefficient with the QFT-ICAM are strong sample-to-sample variations of the path length amplification, as well as fluorescence by pigments that is emitted in a different spectral region than that of chlorophyll-*a*.

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## 1. Introduction

The ability to accurately determine the light absorption coefficient in the visible spectral domain of any kind of particles in fresh- and seawater (phytoplankton, bacteria, suspended minerals, detrital organic matter) is still restricted by some major obstacles. Among these obstacles are a low concentration of particulate matter in oligotrophic waters and the ability of particles to scatter light and therefore often influencing the measurement of attenuation, making still accurate determination of light absorption effects alone a challenging task. Trüper and Yentsch (1967) made measurements of bacteria by concentrating the particles onto optically diffuse glass fiber filters via filtration and determining the absorbance of the filter by measuring the transmittance in a spectrophotometer when placing the filter close to the detector to reduce light losses by scattering as much as possible [1]. This technique evolved from the opal glass technique of Shibata et al. [2,3] and a membrane filter technique to concentrate particles for optical measurements introduced by Yentsch [4]. Due to the ease of use, Kiefer and SooHoo adopted the principal for measurements of particulate absorption coefficients in seawater [5]. The technique was further improved by determining the correction for rather strong path length amplification in the filter (induced by multiple scattering inside the diffuse filter) and referred to it as the "quantitative filter technique" (QFT) [6-8]. The QFT is very effective in making sensitive measurements even in the most oligotrophic waters [9]. A different optical setup that also takes reflectance measurements into account to compensate for filter-to-filter differences in the sample/filter backscattering was introduced as the Transmittance-Reflectance (T-R) technique by Tassan and Ferrari [10]. Other, more elaborate techniques to determine particulate absorption were developed but are not generally in use [11].

Major sources for uncertainties in determining the particulate absorption coefficient,  $a_p$ , with the QFT are the proper corrections for overall scattering effects and path length amplification, the necessary filtration step, and sample filter handling and storage. Often scattering effects are compensated by an offset correction using the measured signal in the near infrared spectral region, assuming that absorption by natural particles in the infrared is negligible and scattering is wavelength-independent. This is surely not the case in coastal waters [12,13]. QFT measurements are usually conducted in laboratory spectrophotometer. As the use of high-end spectrophotometers in the field or onboard of ships is often not practical, it is then necessary to preserve the samples and transport them to the home laboratory. However, losses of pigments are reported by short-time sample handling after filtration [14] and after long storage of frozen samples [15], in addition to possible losses during improper filtration. This makes preservation and transport of sample an additional source for artifacts and uncertainties.

The precision and accuracy of the QFT is limited by variations in scattering by mainly the filter but as well by the particles collected on the filter. The influence of light scattering on the overall absorbance of the filter can be reduced to a negligible level by using a large integrating cavity attached to a spectrophotometer and placing the sample filter in the center of the cavity [16,17], like it was done before for the same purpose with particle suspensions contained in cuvettes [18]. These kinds of spectrophotometers and integrating sphere attachments are sophisticated and expensive equipment not affordable for many labs. On the other hand, simple integrating cavity absorption meter approaches to measure the particulate absorption coefficient in suspension [19] are not sensitive enough to determine  $a_p$ .

in very oligotrophic waters and are not usable for determining the pigment absorption, as in the typical approach the pigments need to be removed by extraction [20] or bleaching [21].

To overcome some of these restrictions, a small, fiber-optic-based optical system was described for the transmittance method [22] that is portable and can be used in the field [23] such that freezing and storage of samples is avoided. This system uses full spectral white light and spectral photodiode array-based detectors, do not take advantages of placing a filter inside a large integrating cavity to reduce light scattering effects, and do not compensate for expected artifacts due to fluorescence from chlorophyll *a* in phytoplanktonic algae (which is not a distinct signal when monochromatic light of low intensity is used, like in a spectrophotometer). However, some of these problems were identified by Miller et al. [22], who described a number of possible advantages of small portable optical equipment to measure optical properties of water.

It is shown that QFT measurements inside an integrating cavity provide significant advantages regarding precision, sensitivity, and reduction of scattering errors compared to other optical setups [16,17]. Here a portable system with an integrating sphere is described that takes advantage of a measurement setup with the filter placed inside the integrating sphere and of the newest spectral detectors, and that allows automatic compensation of artifacts due to chlorophyll *a* fluorescence effects.

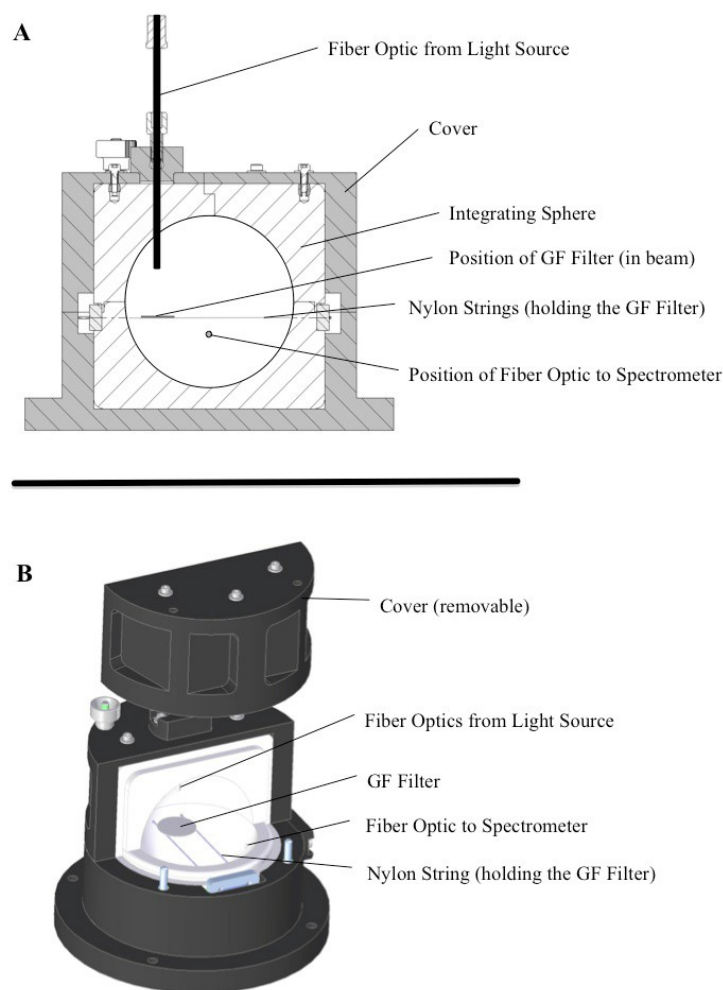


Fig. 1. The Quantitative Filter Technique Integrating Cavity Absorption Meter (QFT-ICAM). A. Schematic view of the integrating cavity (inner diameter 80 mm) showing the positions of the optical fibers and of the GF-filter. Shown here is only the positioning of the GF-filter in the light beam. Moving the GF-filter along the Nylon strings to the right, leads to a position outside the beam. B. An exploded view showing the principal components of the ICAM.

## 2. Materials and methods

### 2.1 Instrument setup

The setup of the integrating cavity absorption meter for QFT measurements, QFT-ICAM (Fig. 1), consists of a spherical cavity made from a highly reflective material, either white PTFE or ODM98 (Gigahertz Optik), with a diameter of 80 mm or 130 mm, respectively, a CF-1000 lamp (Illumination Technology) with an integrated filter wheel as the light source, and a photodiode array detector (SpecSence 2048XL-ULS, Avantes; 300-850 nm, entrance slit: 100  $\mu\text{m}$ ). Optical resolution of the detector is 2.3 nm and spectral resolution is about 0.3 nm.

The three components are connected by two optical fibers in a single beam arrangement. A light beam inside the cavity is arranged with a custom-made, 3 mm in diameter, quartz glass fiber optic ending in a 3 mm thick steel row that is placed from the top into the cavity. The tip of the fiber is about 3-4 cm away from the sample filter. The detector receives light via a 600- $\mu\text{m}$  quartz-glass fiber optic from a hole in the middle of the lower part of the cavity positioned at about 90° towards the light beam (see Fig. 1). Due to the fiber's numerical aperture of  $\text{NA} = 0.22$ , the divergence of the light beam is about 7° and, hence, on the filter an area with a diameter of 8-10 mm is illuminated. Similarly the field of view of the detector fiber is equally determined by the same NA. Due to its positioning inside the cavity, the detector only receives light that is at least once reflected at the walls after being transmitted through and/or scattered by the filter. Therefore no additional baffle is necessary as no light can come directly from the beam to the detectors field of view. The sample filter is held in the middle plane of the cavity by two thin Nylon strings that cross the whole cavity such that the filter can be placed in and outside of the light beam. The 12-position filter wheel inside the light source is equipped with a light-blocking filter made out of aluminum and an optical short-pass filter (SPF; type: 650FL07-25, LOT-Oriel) passing only light with wavelengths <650 nm. The blocking filter is used to measure the detector's dark current signal; the SPF is used to measure the chlorophyll fluorescence signal (around 660 - 750 nm) that is emitted from living algae in the sample.

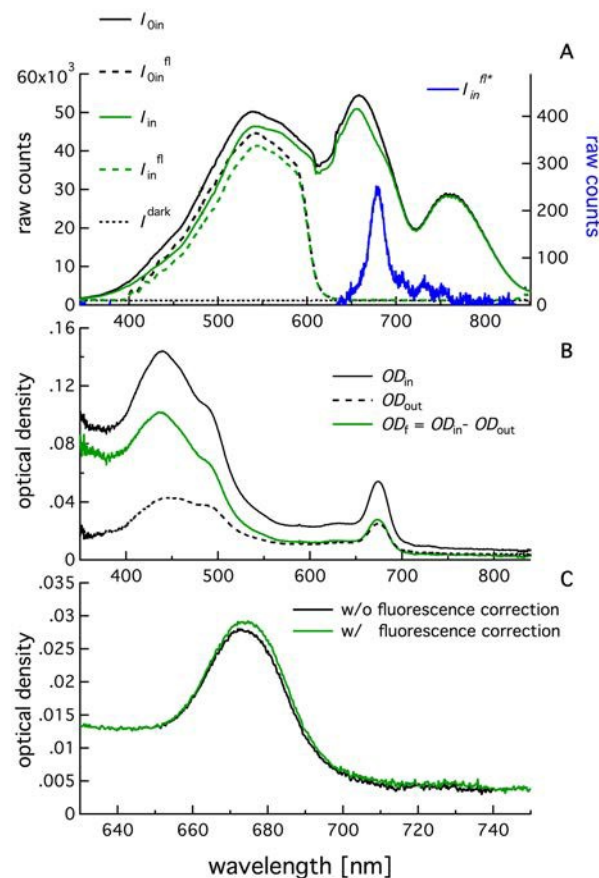


Fig. 2. A. Typical light intensity spectra measured inside the QFT-ICAM when an empty GF-filter (black lines) and when a sample GF-filter (green lines) is placed in the light beam, and when a short-pass filter for chlorophyll fluorescence measurements (SPF; see text) is placed in front of the light source (dotted lines). Additionally shown are the dark current measurements (dashed line) and the calculated intensity from chlorophyll fluorescence emission (blue line, right axis). The legend shows the relevant notations for each spectrum (see text). B. Optical density (OD) spectra calculated from the light intensity measurements when a filter is placed in (black, solid line) and outside the light beam (black, dashed line), and the final OD of the filter sample,  $OD_f$  (green, solid line). C. Calculated OD around the chlorophyll peak at 670 nm with and without a correction for chlorophyll fluorescence.

A top front part of the QFT-ICAM can be removed to place and remove the sample filter (Fig. 1). The detector and the light source are connected via serial/USB cables to a Unix computer. A Python routine with an interactive graphical user interface is used to control both, to initiate the measurement, and to save the results.

## 2.2 Measurement procedure

After the light source had been switched on for more than 10 min to stabilize, a wet, empty glass fiber (GF) filter is placed inside the ICAM's cavity. The integration time for the detector (50 – 300 ms) is set to get a maximum signal of about 85% of the maximum counts of the detector. Typically, 100 - 500 measurements are conducted and averaged; the number of averages is chosen, depending on the integrating time used, to have measurements of one intensity spectrum done in about 10 seconds. A reference measurement is performed by first placing an empty, wetted GF-filter in the light beam and measure the light intensity signal with an open light path (no blocking filter, no SPF). Figure 2(a) shows the different spectral intensity measurements of a typical sample. This first measurement gives the intensity  $I'_{0in}$  (the dependency on light wavelength is omitted here and in the following for simplification). Then, the SPF is placed in front of the light source with help of the filter wheel and a second intensity spectrum is measured to collect the reference for the chlorophyll fluorescence measurement,  $I'^{fl}_{0in}$ . A measurement with the blocking filter in front of the light source is used to determine the dark current,  $I^{dark}$ . Afterwards, the GF-filter is moved outside of the light beam and measurements are conducted to collect the two respective intensity spectra for referencing the optical density signal induced by the diffuse light field inside the ICAM, i.e.,  $I'_{0out}$  and  $I'^{fl}_{0out}$ .  $I^{dark}$  is subtracted from all four light spectra to correct for the dark current of the sensor and, thus, to receive the dark corrected intensities  $I_{0in}$ ,  $I_{0out}$ ,  $I^{fl}_{0in}$ , and  $I^{fl}_{0out}$ , e.g.:

$$I_{0in} = I'_{0in} - I^{dark}$$

The same (five) measurements are made when a sample GF-filter is placed inside the cavity, giving the four dark-corrected intensity spectra  $I_{in}$ ,  $I_{out}$ ,  $I^{fl}_{in}$ , and  $I^{fl}_{out}$ .

## 2.3 Chlorophyll fluorescence correction

Before the spectra of optical density,  $OD$ , of the sample filter can be calculated, the light intensities measured with the sample filter inside have to be corrected for additional emitted light from chlorophyll fluorescence that is excited by the polychromatic light inside the cavity. The procedure is the same as applied for PSICAM measurements [19] and described here in full detail. Chlorophyll fluorescence is emitted in the wavelength range of 650 - 750 nm (see Fig. 2(a)). As for the PSICAM fluorescence correction, any other fluorescence (e.g., from phycobiliproteins) outside of this range is not corrected and absorption spectra of phytoplankton algae containing pigments that fluoresce in other spectral regions might exhibit (partly strong) underestimations of the  $OD$  at those specific wavelengths. The real fluorescence intensities under the full light spectrum (i.e., without the SPF) for a sample filter in- and outside the light beam,  $I^{fl*}_{in}$  and  $I^{fl*}_{out}$ , have to be determined and subtracted from the measured light intensities  $I_{in}$  and  $I_{out}$ . Corrections have to be made due to the facts that 1) some of the fluorescence light is re-absorbed by the sample, 2) the SPF reduces and spectrally changes the excitation, and 3) fluorescence is mainly a function of light absorbed by the pigments in light harvesting complexes of photosystem II, not purely function of light illumination alone.

First: the fluorescence re-absorption is estimated for the situation when the SPF is placed before the light source, assuming that absorption of the fluorescence light is the same as that for light coming from the light source. Therefore the absorption of the fluorescence light is estimated using the "transmittance",  $T$ , calculated from the direct (i.e., not fluorescence corrected) measurements without the SPF, i.e.,  $T_{in} = I_{in}/I_{0in}$  and  $T_{out} = I_{out}/I_{0out}$ . The real fluorescence is calculated at wavelengths between 640 and 750 nm (Fig. 2(a)) for both situations (in and outside the light beam; for clarity the position of the GF-filter is omitted here and in the following) as:

$$I^{fl*} = \frac{I^{fl}}{T} - I^{fl}_0 \quad (2)$$

Second: the difference in total light absorbed by the pigments for the situation with and without the SPF is estimated by using the ratio of all light absorbed by the sample in these two cases. As with an ICAM setup the attenuation of light is mainly by absorption (not scattering), the difference between the light intensity with and without a sample is a good estimate for the light absorption (note that it is ignored here that light absorption for in vivo chlorophyll fluorescence is a linear function of the number of photons absorbed not simply light energy absorbed). The ratio,  $r$ , is, hence:

$$r = \frac{I - I_0}{I^f - I_0^f} \quad (3)$$

Equations (2) and (3) are combined and used to calculate the correct light intensity,  $I^*$ , for the sample filter as:

$$I^* = I - rI^f, \quad (4)$$

for 640 - 750 nm, outside this wavelength range the correction term ( $I_0^f$ ) is set to 0.

The situation for fluorescence is still simplified by assuming that light absorption is by phytoplanktonic algae only. However, the data presented below showed negligible differences at the relevant (i.e., chlorophyll fluorescence influenced) wavelengths between QFT-ICAM and spectrophotometer results, whereas ignoring the chlorophyll fluorescence effect leads to underestimations of the absorption at these wavelengths by up to 10%. In the example shown in Fig. 2(c) the effect is maximally 8%. Because the proposed maximum OD for the here applied measurements should be below 0.2, i.e., the reduction of light intensity by the sample is <22%, the effect will never be larger than 10%. The effect will be stronger when OD is higher. Note that artifacts due to fluorescence by phytobiliproteins can be much stronger at wavelengths where the pigment absorption coefficient is very low, e.g., at 550 - 640 nm.

#### 2.4 Optical density calculation

For both optical situations (in and outside the light beam), OD is now calculated using the real sample light intensity as:

$$OD_x = -\log_{10}(I^*_x/I_{0x}), \quad (5)$$

where the subscript  $_x$  stands for either *in* or *out*. To get the OD of the filter sample for only the first transmitted light,  $OD_f$ , the contribution from the diffuse light field is subtracted (Fig. 2(b)), i.e.:

$$OD_f = OD_{in} - OD_{out} \quad (6)$$

The particulate absorption coefficient,  $a_p$  ( $m^{-1}$ ), is calculated from  $OD_f$  using the obtained filter free area,  $A$  ( $m^2$ ), the path length amplification factor ( $\beta$ ), and the filtered sample volume,  $V$  ( $m^3$ ), as:

$$a_p = 2.303 \cdot OD_f \cdot A / (V\beta), \quad (7)$$

where  $2.303 = \log_e(10)$  and  $A$  is calculated as a circular area using the diameter of the sample patch on the filter; the diameter is regularly measured with a caliber rule. After each sample measurement, the reference measurement with a wet, empty filter is repeated to control stability. The typical short-term stability of full light intensity of the light source is  $\pm 0.1\%$ , however the baseline can change due to small changes in the optical set up, e.g., when manipulating the fiber optics. The resulting two  $OD_f$  spectra for each sample filter measurement were averaged to correct for such small light intensity changes occurring during the time of these three measurements. Typically, the difference in these two  $OD_f$  spectra is below the precision for replicate measurements (see below). It was made sure that the maximum  $OD_f$  was in the range of 0.08 to 0.2.

#### 2.5 Filtration and bleaching

Filtration of a specific sample volume (0.05 to 4.5 L) is done onto combusted 25- or 47-mm GF-filters (Macherey-Nagel type GF-5 or Whatman type GF/F) using a 100-ml or 250-ml glass filtration unit (Sartorius) and a 2-L glass vacuum bottle under low vacuum of <100 hPa. The sample filter is stored at room temperature in a closed petri-slide on a dark place for maximally several minutes. Large filters are cut into four quarters. One quarter is checked for wetness by placing it shortly on a clean white tissue, if too dry (when no wetting of the tissue is observed) it is moistened by placing it on a few drops of filtered seawater. Due to the short time after filtration, often no extra moistening of the filter was necessary. The filter quarter is directly placed inside the ICAM for measurement. Typically, the procedure for one measurement took about 2 minutes. In case 25-mm sample filters are used, they are prepared accordingly and as a whole placed inside the cavity. If the maximum  $OD_f$  is outside of the desired range (see above), another filter is prepared

by filtering an adequate (i.e., smaller or larger) sample volume.

After this first measurement, each filter is bleached by putting it for a few minutes onto a few drops of a 10%-NaOCl solution [10,21]. The filter is then placed onto a dry tissue to remove most of the free bleach and re-wetted with some drops of filtered seawater. The bleached filters are measured as described above, except that no correction for the chlorophyll fluorescence is necessary, so, measurements with the SPF in front of the light source are omitted for bleached filter to shorten the measurement procedure. Typically the whole measurements procedure for one sample took 15-20 min, including several filtrations, several sample and reference measurements, and bleaching.

## 2.6 PSICAM measurements

Measurements with a point-source integrating-cavity absorption meter (PSICAM) are used to determine the absolute absorption coefficient of the particulate absorption for comparison and to individually determine the path length amplification factor. The measurements are performed as described by Röttgers et al. and Röttgers & Doerffer [19,24]. In short, the absorption of the sample, i.e., the sum of particulate and dissolved matter,  $a_{g+p}$ , is determined with the PSICAM when purified water served as the reference. Purified water was freshly prepared with a Synergy Ultrapure water system (Millipore) that was fed with purified water prepared earlier with a larger purification system (ELIX and Gradient, both Millipore). Temperature and salinity effects on the water absorption are corrected using instrument-specific correction factors [24] and the individually measured temperature and salinity of the sample and the reference. Each sample is measured three times alternating with measurements of the reference, i.e., purified water. The sample is then filtered, first through a combusted GF-filter, then through 0.2  $\mu\text{m}$  (GSWP; Millipore). The absorption of the filtrate,  $a_g$ , is then measured in the PSICAM and, for controlling accuracy of the PSICAM, additionally in a LWCC system (WPI) with an optical path length of 2.5 m. For the LWCC measurements purified water served as the reference, the temperature of the sample and the reference was kept the same, and the optical effect of the difference in salinity were corrected with a salinity correction spectrum that is determined from measurements of a 100 g/L (taken to be equal to 100 PSU) NaCl solution. Measurements of colored solutions (made from Nigrosine) are used to calibrate the PSICAM. The difference of these two absorption determinations gives the particulate absorption,  $a_p = a_{g+p} - a_g$ .

## 3. Assessment

### 3.1 Determination of measurement precision

In a first test the repeatability of measurements with the QFT-ICAM is examined by measuring several times ( $n=11$ ) the same wetted filter and different wetted filters of the same batch against air to determine the precision. This had been done for comparable optical setups by Röttgers & Gehnke [16]. In that study the precision (i.e., standard deviation) for  $OD_f$  measurements inside an integrating sphere was determined for different aspects (wetting of filters, variations in a batch of filters, etc.) and ranged (over wavelength) between 0.0001 and 0.0010, with mean values (average over all wavelengths) for the different aspects of 0.0003 - 0.0008 on the OD level. The same values for the precision of transmission measurements made in front of an integrating sphere were more than one order of magnitude higher and varied between 0.0015 and 0.0120, with mean values between 0.0022 - 0.04. As the QFT-ICAM is designed to be used in the field and onboard ships, it was compared here with a QFT2-filter holder (WPI) for transmittance measurements that can optically be connected to the same light source and detector [see 22]. When measuring several filters of one batch in the QFT-ICAM, the precision for  $OD_f$  ranged between 0.0002 and 0.0022 over all wavelengths (350 - 750 nm) (Fig. 3), the mean value was 0.0006. The values for measuring repetitively the same filter were similar (data not shown). The absolute precision is a function of the light intensity inside the cavity, i.e., higher values are observed in regions of low light intensity (e.g., <400 nm). The values for repetitive transmittance measurements with the QFT2-filter holder were more than one order of magnitude higher and ranged from 0.0064 to 0.0095 with a mean value of 0.0076, and, hence, showed about the same precision as found by Röttgers and Gehnke for transmittance measurements with a spectrophotometer [16]. The attainable precision for the simple setups (QFT-ICAM and QFT2 filter holder) is the same as that for high-end spectrophotometers.



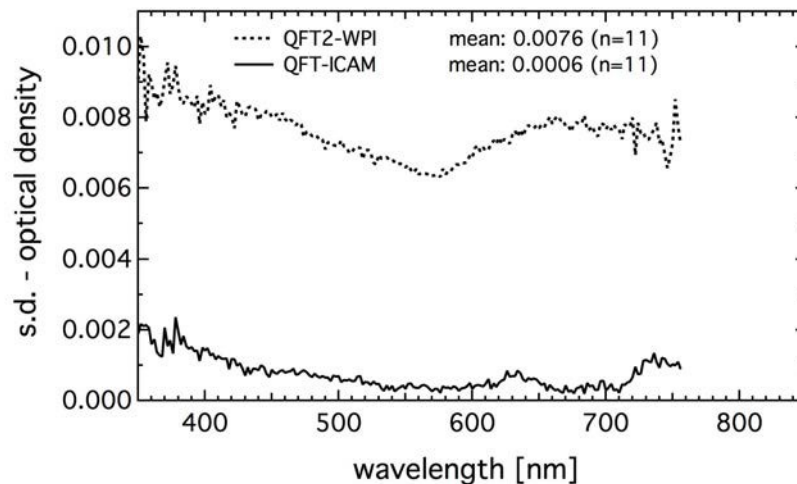


Fig. 3. Standard deviation (s.d.) for repetitive optical density measurements of several filters in the QFT-ICAM and with the QFT2 filter holder. Shown is s.d. as a function of light wavelength.

### 3.2 Estimation of accuracy

Miller et al. (2011) showed that a portable setup with the QFT2-filter holder provides similar results for transmittance measurements to those in a commercial spectrophotometer, except a visible artifact due to the influence of chlorophyll fluorescence on the measurements with the portable filter holder setup [22]. The reason for this fluorescence effect is the use of full spectral light and a hyperspectral detector. It was therefore assumed that the QFT2 filter holder setup behaves similar to the spectrophotometer setup for simple transmittance measurements and its performance with sample filters was not further investigated. The advantages of the integrating sphere setup can clearly be seen in the much better precision (Fig. 3). A direct comparison of the QFT-ICAM with the QFT2-filter holder was, hence, not conducted; instead measurements with the QFT-ICAM were compared to measurements in a lab spectrophotometer following the spectrophotometric method for inside integrating sphere QFT-measurements [16].

High-end spectrophotometers do normally provide measurements with an accuracy of 0.0003-0.0005 OD (e.g., Lambda 950, Perkin-Elmer, double-aperture method for 0.5 OD, accuracy: 0.0003, precision: 0.0008). However, with the QFT method, the accuracy for the OD determination is dependent as well on the accuracy of the determination of the path length amplification factor. The accuracy of the instrument (mainly determined by the quality of the compensation of the detector's non-linearity) is often controlled by using certified neutral density filters of known OD. This was not possible for the QFT-ICAM as the size of the light spot is larger than the dimensions of the available NIST-traceable neutral density filters. Therefore, to estimate the overall accuracy of the QFT-ICAM, measurements with larger (50 x 50 mm), uncertified neutral density (ND) glass filter were performed in the QFT-ICAM and a recently calibrated Lambda 950 double-beam spectrophotometer (Perkin Elmer) equipped with a 150-mm integrating sphere. Each filter was placed in the center of the specific integrating sphere of each setup. Measurements were performed for the same wavelength range, i.e., 350 - 850 nm, with a spectral and optical resolution of 2 nm (slit width for 2 nm) and a scan speed of 100 nm/min for the spectrophotometer. Two different ND filters were used that had mean OD in the visible spectrum in the typical range of measurements, i.e., 0.131 and 0.273, respectively. Before a comparison can be made, the technical difference between a double-beam and a single-beam setup needs to be taken into account.

In a double-beam spectrophotometer the output signal is related to the difference in the electronic level between the detector's signal induced by the sample beam and that induced by the reference beam. A single light beam from the light source is split into a sample and reference optical channel. With a chopper system the two channels are illuminated alternately onto the same PMT detector. When performing an "autozero", deviations between the PMT signals of the two channels/beams are electronically compensated, such that the output difference under these conditions is set to zero (i.e., transmittance is 100%). Variations of the intensity of the light source over time do not change this output signal as they do not change the relative differences between the two beams, which makes a double beam instrument more stable than a single beam one. With a large integrating sphere attached, both light beams do enter the sphere through two entrance ports. The sample is placed inside the cavity in the sample beam but outside of the path of the direct reference beam, the reference beam typically passes behind the sample. With the highly diffusive filter type used here, the sample beam is scattered by the filter and only a small portion of the beam will directly reach the sample beam's exit port. This scattered light when reflected by the walls of the cavity, thereby getting more and more diffuse, can interact with the sample another time or even several times. The reference beam passes the sample and is diffusively reflected by a highly reflective material of a white standard plate that closes each exit port. After this first reflection, the

light of the reference beam behaves similar to that of the sample beam, i.e., the light intensity reduction from additional interactions of the sample beam with the sample is automatically compensated by the same kind of interactions of the reference beam with the sample. The final signal is, hence, just the light intensity reduction by the first interaction of the sample beam with the sample, additionally influenced by the difference in reflectance of the sample filter and the white reflection standard plate, this difference is compensated by performing baseline measurements with empty filters.

The QFT-ICAM is a single-beam instrument. For compensation of the effects of additional interactions of the diffuse light that is produced by the light beam after interacting with the sample for the first time, the OD is measured in two situations (see above), once when the sample is illuminated directly by the light beam and once when the sample is placed outside the beam. The assumption here is that the diffuse light field inside the cavity does not differ by whether the direct beam is diffusively scattered by the GF-filter or by the cavity wall (when there is no GF-filter in the beam). Compared to a double-beam spectrophotometer setup, the rather simple QFT-ICAM setup is considered to be i. less effective in compensating for the above effect, ii. as a single beam instrument less stable, and iii. less accurate due to a larger divergence of the light beam. However, by using a non-scanning, full spectral approach, averaging several detector readings for the whole light spectrum is done much faster, and the electronically stabilized light source is stable enough to perform measurements for a period of several minutes. As reference measurements can be done within the same short time, repeated reference measurements are used to compensate for longer-term intensity variations of the light source. For the direct comparison it was made sure that the overall time used for each sample measurement was roughly the same, i.e., the number of averages used with the QFT-ICAM was increased to fit to the full scanning time of the spectrophotometer. This reduced much the expected stronger noise in the QFT-ICAM measurements.

Each of the two ND filters was measured three times in the spectrophotometer, with air readings in between, after the baseline was set with air. To fix the filter in the center of the integrating sphere, a clamp-style filter holder was used. The filter was based on the basal baffle of the holder and fixed with the clamp. In the QFT-ICAM the filter was regularly placed onto the two nylon strings and measured three times in both positions (in and outside the beam) with reference air measurements in between. For the spectrophotometer results, the three respective readings were averaged and the mean air reading subtracted from the mean filter reading. The OD signal of air (compared to the former baseline) was below 0.001 in the visible (400 - 700 nm). Standard deviations, *s.d.*, for these two sets of three spectra (filter and air) were calculated for each filter and simply added for error propagation. From the three QFT-ICAM filter measurements, six OD spectra were calculated for each filter spectrum, one with the air spectrum measured before and one with that measured afterwards. The six spectra were averaged, and the *s.d.* determined over all six spectra.

For the QFT-ICAM setup, wavelengths with generally low light intensity will have the largest relative uncertainty, as the influences of the detector's internal stray light and non-linearity will be largest. To avoid stronger biases due to these detector errors at low light intensity, the following analysis is limited to values measured at 400 - 800 nm. The *ODf* measured in the QFT-ICAM differed between -1.5% and 0.5% from that of the spectrophotometer, with *s.d.* values of between  $\pm 1.0\%$  and  $\pm 1.5\%$  in both measurement setups. Deviations of about 1.3% to 1.5% were statistically significant (paired t-test,  $p < 0.0005$ ,  $df = 7$ ), i.e., there is a small tendency of the QFT-ICAM underestimating *ODf*, however these are considered as being still small compared to general measurement uncertainties when measuring GF-filter. Also the number of repetitions and general measurements were too small to come to a final conclusion about this small deviation. Hence, the QFT-ICAM can provide measurements with a similar accuracy and precision as a high-end spectrophotometer.

### 3.2.1 Comparison using sample filters

In addition to the uncertainties related to the precision and accuracy in the *ODf* determination, measurements of particles on GF-filters are biased by inhomogeneity of the particle distribution on the filter and variations of the path length amplification effect. Therefore, several samples ( $n = 30$ ) from the HZG filter sample archive were measured with the Lambda 950 spectrophotometer and the QFT-ICAM. These were filters prepared from water samples taken in the German Bight in 2014 that had maximum OD between 0.05 and 0.2, to fit to the desired range for OD. Each sample filter was wetted with some drops of water and measured typically once in the spectrophotometer and once in the QFT-ICAM. When visible stronger deviations between spectrophotometric and QFT-ICAM measurements were observed, the same filter was measured a second and third time in both instruments.

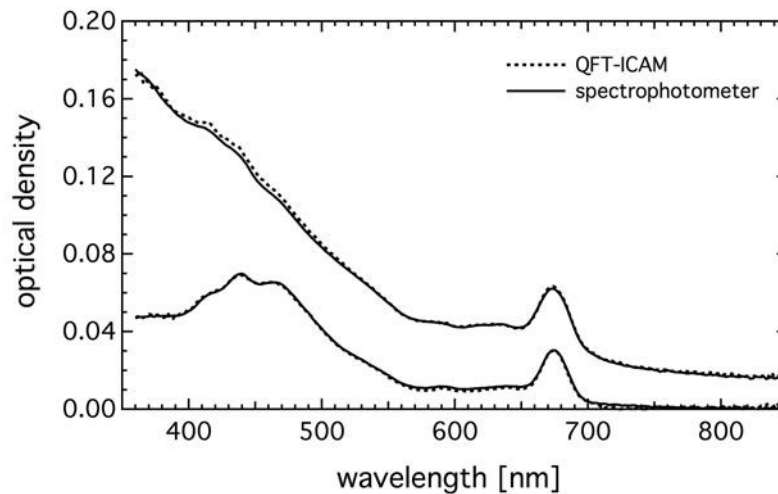


Fig. 4. Optical density as a function of light wavelength for two sample filters. Shown are results for each filter when measured using the QFT-ICAM and when using the Lambda 950 spectrophotometer.

Figure 4 shows two examples of this comparison, for a sample showing a low and for one showing a high contribution of non-algal matter, visible by a low and high absorption coefficient at near infrared wavelengths, respectively. For both samples the two methods gave the same (on the level of precision)  $OD_f$  values even at wavelengths of 650 - 750 nm, indicating that the influence of chlorophyll fluorescence is compensated successfully in these two cases. Several more cases of such a very good agreement are observed. In other cases, the deviations were stronger, typically the differences then showed a wavelength-independent relative bias. The same sample filter is then measured again, showing that the OD values varied rather strongly between repetitive measurements done with both instruments. A closer visual inspection of some of these filters showed a slightly inhomogeneous distribution of the material on the filter. As with both optical setups repetitive measurements cannot be conducted at exactly the same spot on the filter, an inhomogeneous distribution of the material on the filter is a predictable reason for variations in repetitive measurements and, thus, for the differences between the two instruments. Not only the position of the light beam on an individual filter is not the same for each instrument, the area of the spot illuminated by the light beam in both instruments is also not the same; in the spectrophotometer the area illuminated by the beam is rectangular with dimensions of about 3 x 10 mm, whereas in the QFT-ICAM it is a circular, larger spot of about 10 mm in diameter.

The values of the bias induced by inhomogeneous distributions of the particles are expected to vary rather stochastically over all samples. Indeed, a linear regression analysis for this correlation for all wavelengths revealed intercept values close to zero, below the precision. The linear regression was then forced through the origin, to be able to use the deviation of the regression coefficient from 1 as an indicator for comparability of the two instruments. The regression coefficient (slope) varied between 0.95 and 1.00 for the range of 360 - 750 nm, average 0.97, only at >750 it decreased to minimally 0.9. Excluding, consecutively, data of filters showing stronger variation in repetitive measurements did not change the regression coefficient. There seems to be a small general underestimation of OD by the QFT-ICAM compared to the spectrophotometer, by on average 3%. Nevertheless, in most cases and at most wavelengths these 3% were still below the level of precision.

Normalization of the OD spectra of one sample to the underlying area showed that the spectral distribution was always exactly the same, i.e., the deviations in repetitive measurements are mainly wavelength-independent relative differences explainable by different particle concentrations in the light beam due to an inhomogeneous particle distribution. So, in addition to general measurement errors, the level of homogeneity in the distribution of the material on the sample filter and the variation of the path length amplification factor is an additional source for variations in  $OD_f$ . The amplification factor is likely but to an unknown extent dependent on the measuring conditions and can easily explain such small deviations of 3%.

In summary, the two instrumental setups are comparable in determining  $OD_f$  of a sample in terms of precision and accuracy. Any systematic deviations are smaller than uncertainties explainable by typical determination errors related to the variation in path length amplification and the inhomogeneity in the distribution of material on the filter.

### 3.3 Field samples

Prototypes of the QFT-ICAM were tested during two field campaigns in waters of the New Caledonian Lagoon during a ship cruise with the RV *L'Alis* in April 2014 and in waters around the British Isles during a ship cruise with RV *Heincke* in April 2015. A total of 125 water samples are taken from which about 300 filter samples are prepared. Therefore, depending on the particle concentration and the size of the filter used, between 20 mL and 4.5 L of the sample water are filtered onto GF filters. The samples are immediately measured onboard using the QFT-ICAM. In parallel, determinations of the particulate absorption in suspension were conducted with a PSICAM. During the April '14 cruise, 47-mm GF-5 (Macherey & Nagel) are used, whereas during the April '15 cruise 25-mm GF/F (Whatman) filters are used for measurements in the QFT-ICAM. During the cruise in April '15, duplicate 25-mm filters are prepared for QFT-ICAM measurements and additional filters are prepared on 47-mm GF-5. The latter are flash-frozen in liquid nitrogen, stored at  $-80^{\circ}\text{C}$ , and measured with the Lambda 950 spectrophotometer setup two weeks after the cruise. A total number of 312 samples filter samples are collected with maximal  $OD_f$  values  $<0.2$ . During the cruise in April '14, very low absorbing waters were encountered. The subsamples for the determination of the absorption coefficient by the dissolved fraction, from  $0.2\text{-}\mu\text{m}$  filtered samples, with the PSICAM were observed to be contaminated occasionally, i.e., having an absorption coefficient in the blue that was higher than the values of the respective unfiltered sample. Additional measurements of the filtered samples with a liquid waveguide system done before the PSICAM measurements showed less optical contamination and were, hence, used instead. However, in a few cases the particulate absorption coefficient was too low to get good results with the PSICAM.

Based on the PSICAM results, the water samples comprised a large variety of absorption coefficients from very oligotrophic water outside the New Caledonia Lagoon to very turbid waters close to the British coast. The particulate absorption coefficient varied over three orders of magnitude, between  $0.008$  and  $2.6\text{ m}^{-1}$  at  $442\text{ nm}$ , and between  $0.003$  and  $0.9\text{ m}^{-1}$  at  $672\text{ nm}$ . Spectral behavior of the particulate absorption showed phytoplankton dominated as well as non-algal material dominated particle assemblages and all kinds of mixtures of algal and non-algal material.

It is shown above that both optical methods (QFT-ICAM and spectrophotometer setup) are comparable in terms of precision and accuracy, the focus here is on effects by freezing and storing samples and the variations of the path length amplification factor. This comparison and the individual determination of the amplification factor were possible by the additional determination of the particulate absorption with the PSICAM that here serve as a standard method for accurate determination of the particulate absorption coefficient. Note again, that PSICAM measurements are limited to a narrower wavelength range, to situations when the maximum absorption coefficient is  $>0.01\text{ m}^{-1}$ , and that they are done with particles in suspension, hence, cannot be used to determine the non-algal matter absorption alone.

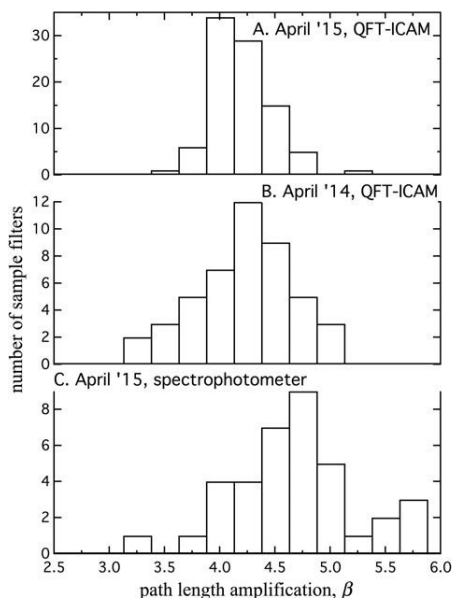


Fig. 5. Histograms of the path length amplification factors,  $\beta$ , for different data sets. A. Direct QFT-ICAM measurements during April '15 in waters around the British Isles ( $n=93$ ). B. Direct QFT-ICAM measurements during April '14 in waters of the New Caledonian lagoon ( $n=46$ ). C. Separate spectrophotometer measurements of the April '15 samples, done 14 days after the end of the cruise ( $n=37$ ).

#### 3.3.1 Path length amplification

Measurements of  $a_p$  with a PSICAM have been used earlier to determine the path length amplification [16]. These results showed good linear correlations between  $OD_f$  and the true particulate absorption coefficient, if maximum  $OD_f$  of a filter was below 0.1. When  $OD_f$  was higher, this relationship became more and more non-linear, due to particle self-shading and/or when particle scattering dominates filter scattering and thereby alters the backscattering of the sample filter. Often this non-linearity in the  $OD_f$  vs.  $a_p$ -relationship is corrected by fitting a non-linear (e.g., a quadratic) function to the data [8]. When correlating values of  $a_p$  calculated directly from the  $OD_f$  with the real  $a_p$ , the resulting function or slope (in case of a linear function) represents the factor of the optical path length amplification inside the GF-filter,  $\beta$ . The result of the reverse correlation is the path length amplification correction factor. Taking a linear approach, Röttgers and Gehnke determined amplification factors for about 20 algal culture samples in the range of 3.5 to 5.4 with a mean value of 4.50 and for about 30 natural samples of 3.8 - 5.1, mean 4.47 [16]. Stramski et al. presented another correction for QFT measurements inside an integrating sphere [17]. Correction functions for the path length amplification was determined from measurements of the particles in suspension and on the filter made inside the same integrating sphere. As done in most previous work, non-linear function to calculate OD in suspension,  $OD_s$ , from  $OD_f$  are presented. Note, that this is not possible with PSICAM measurements, as the PSICAM is not based on a beam attenuation principal, and no  $OD_s$  is measured, it can only be theoretically calculated from  $a_p$  for a given path length. The comparison is not easily made when non-linear function are used; with the power function presented in [17], the amplification factor for an  $OD_f$  of 0.01 and 0.2 are about 4.6 and 3.6, respectively.

Before analyzing  $\beta$  values in detail, a data quality analysis is performed, with the assumption that the real  $a_p$  and  $OD_f$  are linearly related when  $OD_f$  is below 0.2 as was shown to be a valid hypothesis [16]. A non-linear (quadratic) relationship appears when  $OD_f$  values are reaching levels that self-shading by particles on the filter gets significant, or when wavelength-dependent errors occur in either the  $OD_f$  or the independent  $a_p$  measurement. In the data set used here this kind of error occurred due to optical contamination of the absorption coefficient measurement of the dissolved fraction with the PSICAM. Such an error would lead to an underestimation of  $a_p$  at wavelengths where the dissolved fraction absorbs strongly. The overall effect will depend on the level of contamination and the ratio of  $a_p$  to the absorption coefficient of the dissolved fraction. It would lead to a non-linear behavior of  $\beta$  over the wavelength range, actually in the opposite way than particle self-shading. Each error in each step of the  $a_p$  and  $OD_f$  determination has a direct effect on  $\beta$  and its behavior with wavelength (and thus  $OD_f$ ). As the data from field measurements presented here are partly from samples with very low  $a_p$  values, the PSICAM measurements were often close to the detection limits and some spectra showed significant noise. Single spectra showed visible artifacts consistent with relative large measurement uncertainties, hence, in a first step, linear regressions of  $a_p$  vs. OD are performed for all 312 data sets. All data sets for which the coefficient of determination,  $r^2$ , was below 0.95 were omitted from the following analysis ( $n=17$ ). Besides visible spectral artifacts or a general non-linear relationship in the correlation,  $r^2$  is lower when a larger level of noise is apparent in one of the spectra (here in  $a_p$ ). To analyze whether noise or a non-linear relationship leads to a lower  $r^2$  value of the linear regression analysis, a quadratic regression was performed for each data set. Typically, due to the higher degree of freedom in the fit of a quadratic function (second order polynomial), the  $r^2$  value is closer to 1 than that of a linear regression analysis. With general noise in the data, this increase in  $r^2$  for the quadratic regression analysis is low, but stronger when the relationship is non-linear. In addition, the value of the quadratic term of the regression indicates whether  $\beta$  increases or decreases with  $OD_f$ . The number of data sets for which this quadratic term was positive and that for which it was negative is about the same, showing that any non-linear behavior in this relationship for the samples used here most likely results from wavelength-dependent measurement errors not from a general non-linear behavior induced by particle self-shading. In most cases, the term was close to zero indicating a good linear relationship. To avoid this kind of measurement errors influencing the statistical analysis of the  $\beta$  factor, the number of data sets was further reduced by choosing only those data sets for which  $r^2$  of the linear regression analysis was  $>0.995$  ( $n=174$ ).

The data sets were analyzed for the mean  $\beta$  factor. This analysis was done separately for the two cruises and for samples measured with the QFT-ICAM and those measured later in the lab with the spectrophotometer. The mean ( $\pm$ s.d.)  $\beta$  factors are  $4.09 \pm 0.44$  (April'14, QFT- ICAM, 47-mm GF-5,  $n=46$ ),  $4.06 \pm 0.28$  (April'15, QFT-ICAM, 25-mm GF/F,  $n=93$ ) and  $4.56 \pm 0.54$  (April'15, Lambda950, 47-mm GF-5,  $n=37$ ). The latter value is significantly different from the first two values ( $p<0.01$ , ANOVA). Histograms of  $\beta$  for the three sets of data are shown in Fig. 5. The above assessment has shown that differences between QFT- ICAM and the spectrophotometer setup are low for the same sample filter and the results of the mean  $\beta$  factor showed that the type of filters (25-mm GF/F vs 47-mm GF-5) has no influence on the mean  $\beta$  factor. The significant differences of the measurements done later in the lab on frozen samples might indicate that the sample treatment (freezing and storing) is affecting path length amplification. Several possible influences on the path length amplification are shortly tested, including soaking, freezing, and drying of the filter, as well as the influences of the type of filter material (data not shown). No visible

differences are observed, except when filters were completely dry, this increases the OD by several percent. At the moment, the differences in  $\beta$  observed here between data sets are without explanation.

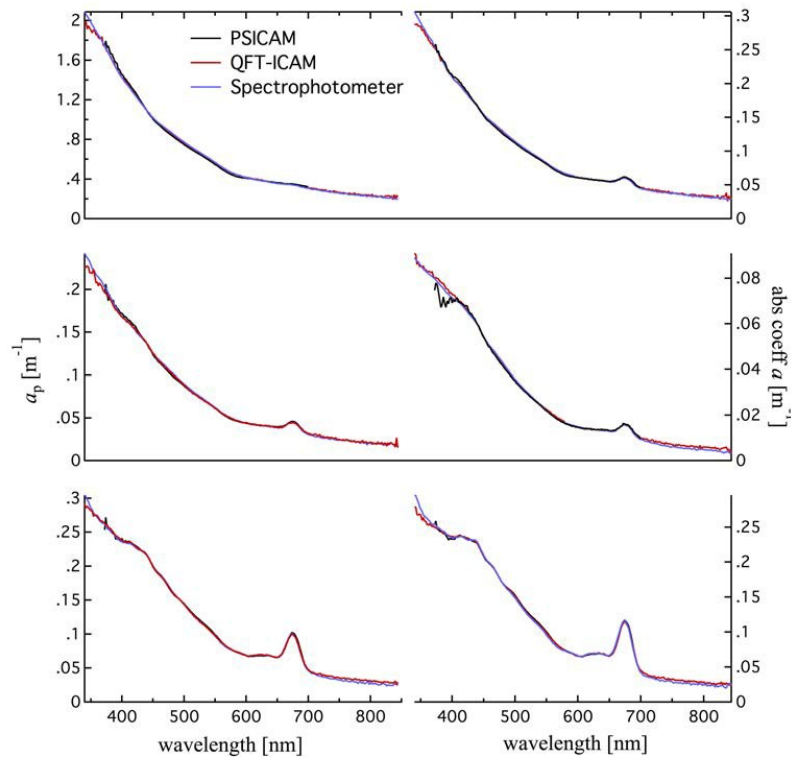


Fig. 6. Comparison of particulate absorption spectra determined from QFT measurements inside an integrating sphere. Shown are results of QFT-ICAM measurements done directly after sample filtration onboard a research vessel and spectrophotometric measurements later in the home lab done with filters of the same sample that had been frozen and stored. For comparison, results of PSICAM measurements on the original sample (particles in suspensions) are shown. Note, the absolute absorption coefficients of each sample are adjusted to be the same by using the PSICAM results to determine a mean path length amplification correction factor for each filter for wavelengths of 400-500 nm and 600-700 nm, the 500-600 nm is ignored due to artifacts induced by fluorescence of phycobiliproteins in some samples.

For the sample set with the lowest variability (April '15, QFT-ICAM) duplicate sample filters are prepared. Differences in each duplicate are considered being induced by an inhomogeneous distribution of the particles on one filter or on both filters of the duplicate. That this is a significant error source in determining the real  $\beta$  factor can be derived from variations in  $OD_f$  when comparing single filters as described above. The maximum difference of  $\beta$  in all duplicates ( $n=63$ ) was 12.6 %, and in 24% of the samples it was  $>5.0\%$ . Inhomogeneity in the particle distribution can, hence, partly be responsible for the observed variations in the  $\beta$  factor.

### 3.3.2 Effects of filter freezing and storage

An analysis of individual samples of the April '15 cruise for spectral artifacts is done by using for each sample filter individually determined  $\beta$  factor and comparing the resulting absorption coefficient spectrum with that obtained from suspension measurements (with the PSICAM). As it will be shown here, small pigment losses can be observed at around 550 nm in some samples. The visibility of these losses was enhanced by omitting the spectral region of 500-600 nm from the linear regression analysis to get the correct  $\beta$  factor.

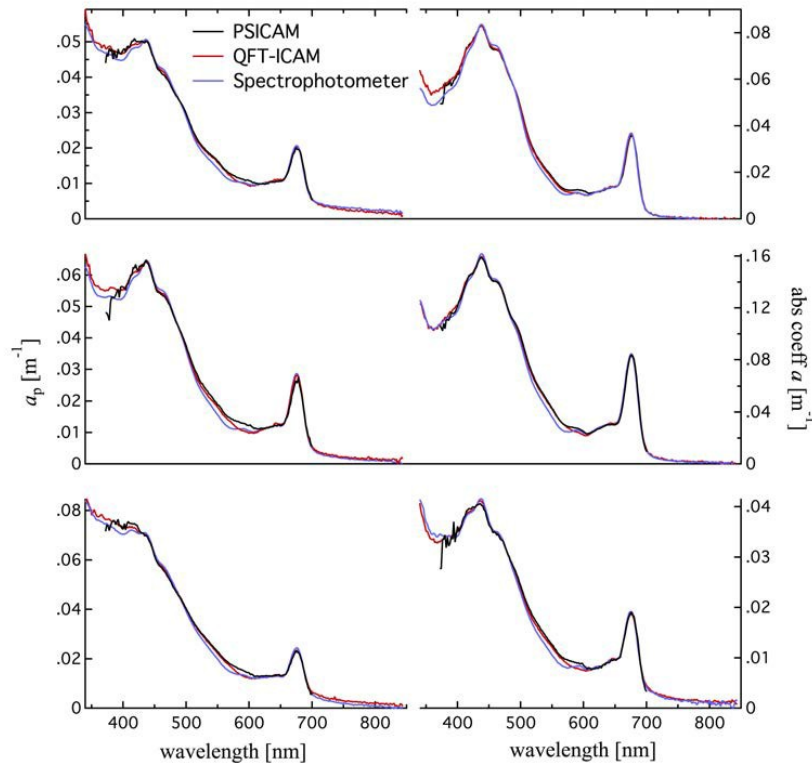


Fig. 7. Same as Fig. 6, but for six samples with visible differences between direct and later QFT measurements.

Per sample two filter spectra are obtained, one from a filter measurement in the QFT- ICAM and one from a filter that was frozen, stored, and measured later in the lab. In the ideal case, these two spectra are identical over the full wavelength range and spectrally fit well to the PSICAM results. Well-matching spectra are observed for the majority of the samples (Fig. 6). However, this majority consisted of samples which absorption is dominated by non-algal matter. When particulate absorption is dominated by phytoplankton, lower absorption coefficient values were observed consistently at 500-600 nm in frozen and stored filters (Fig. 7), whereas the QFT-ICAM spectra fit well to the PSICAM spectra (except at ca. 580 nm, see later). The spectral range of this lower absorption indicates losses of pigments during freezing or storage that absorb in this specific spectral region, most probably phycoerythrin, a chromophore of water-soluble phycobiliproteins. When inspecting the spectral results of the QFT-ICAM measurements in more detail, lower absorption coefficients (compared to PSICAM results) are observed at around 570-600 nm (Fig. 8), the wavelengths of phycoerythrin fluorescence. The appearance of such fluorescence would lead to underestimations of the absorption coefficient at these wavelengths. In principal, the same effect should be observable in PSICAM measurements. For a very low absorbing sample (Fig. 8), a higher OD is measured in the QFT-ICAM (concentrated particles on a filter) than in the PSICAM (original particle suspension), i.e., a much higher absolute light intensity difference is measured between sample and reference. The effect of the induced fluorescence light intensity on the absorption coefficient depends directly on the ratio of fluorescence light intensity to light intensity reduction by the sample, hence, in the here obtained cases of low absorbing water, it will be stronger for QFT-ICAM measurements.



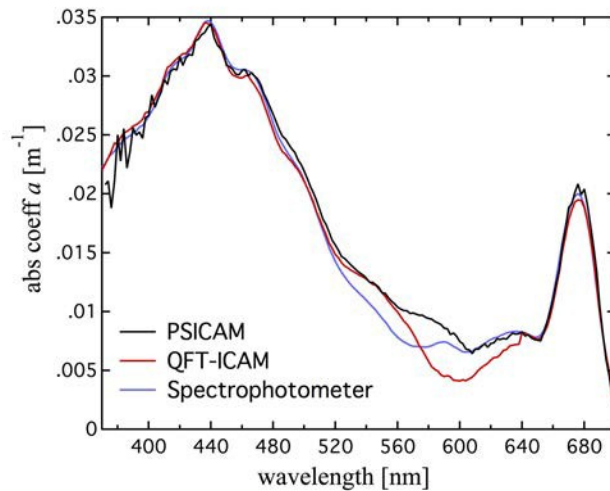


Fig. 8. Same as in Fig. 7 but for a sample with very low particulate absorption coefficient, to highlight the spectral differences at 500 - 640 nm.

Based on the strength of the absorption shoulder at 550 nm, the concentrations of phycoerythrin in these samples were very low, the losses are, hence, not well pronounced but measurable. The effects of these losses are as well visible as small hystereses in the correlation plots of the absorption coefficient obtained from PSICAM and QFT measurements (data not shown). The results still indicate that these pigments are lost during freezing and storing of samples, but not during careful filtration. These losses might be stronger for samples with higher concentrations of phycobiliproteins. Problems for frozen filters with water-soluble pigments (mycosporine-like amino acids) in the ultraviolet spectral region have been reported earlier [15,25]. The results presented here indicate also that losses of regular, water-insoluble pigments by filtration, freezing and storing (for up to 30 days) are very low and did not lead to a measurable spectral effect on the absorption coefficient [see 14,15].

#### 4. Discussion

A compact and portable spectrophotometric setup (QFT-ICAM) is presented that allows measurements of GF filters with aquatic particles inside an integrating sphere for accurate determination of the particulate absorption coefficients in natural waters. The setup provides several advantages when conducting these measurements in the field or onboard of research vessels, when, due to their size and costs, high-end spectrophotometers cannot be used. Miller et al. outlined these advantages during the assessment of the WPI-QFT2 filter holder [22]. As already applied with other optical configurations [16,17,19], the filter is measured inside an integrating sphere to reduce scattering errors and a correction of chlorophyll-*a* fluorescence effects is implemented here. However, fluorescence light from phycobiliproteins is affecting QFT-ICAM measurements and needs to be corrected when measuring samples with phycobiliprotein-containing phytoplanktonic algae. (A similar approach as done for the influence of chlorophyll fluorescence might be feasible, using a <550 nm short-pass filter.)

It is conclusively shown that the obtained precision with the QFT-ICAM is much better than for filter measurements without an integrating sphere. The precision is much lower for the QFT-ICAM, but very similar to that of the same ICAM setup when using a double-beam spectrophotometer [16]. The similar precision for filter measurement with the QFT-ICAM and the high-end spectrophotometer indicates that scattering effects induced by variations in optical properties of GF-filter are the main cause in determining the precision, whereas instrument-specific characteristics (electronic noise, detector's non-linearity, etc.) play a minor role. It is therefore not surprising that such a simple setup can provide very accurate measurements that lay not behind that of more sophisticated instrumentation.

The strong reduction of filter scattering effects with this setup had been shown to allow accurate determination of  $a_p$  in the infrared spectral region [16], making an often applied offset correction to adjust for scattering effect obsolete. The good agreement of QFT-ICAM and measurements with a commercial spectrophotometer, also in the infrared region, confirmed that this offset correction is never necessary for inside integrating sphere measurements. It will provide more accurate measurements in coastal areas, where infrared absorption by non-algal matter can be very high.



With the possibility of concentrating particles on a filter, the QFT technique allows incomparable sensitivity in determining  $a_p$ , even in the most oligotrophic waters. The high path length amplification for filters inside an integrating sphere adds another advantage with respect to sensitivity and filtration/sampling effort. The fast execution of measurement allows prompt adjustment of the sample's filtration volume for optimization of the filter's  $OD$ . It is shown here that this method, without the need of sample preservation, can avoid artifacts due to pigment losses or degradation during the otherwise necessary freezing and storing procedures.

Besides a possible correction for phycobiliprotein fluorescence effects in QFT-ICAM measurements, a consistently remaining problem of the QFT-technique that limits accurate  $a_p$  determination is the apparent variability in path length amplification [16,17] in either optical setup. The variations over the mean of one data set exceeds  $\pm 20\%$  (2xs.d.), with an additional  $\pm 10\%$  variation of the mean between data set, with some single extreme values for the QFT- ICAM (min: 3.2 and max: 5.7). These extreme values are quite rare, and probably due to some procedural errors, e.g. an inhomogeneity of the particle distribution in the filter can easily induce  $\pm 10\%$  variation of the  $OD_f$ , and, hence, seemingly, of the path length amplification. Avoiding or analyzing inhomogeneity in particle distribution might reduce variation in  $\beta$ , however, other parameters, like cell size, are considered to have effects on the  $\beta$  factor. The presented data showed that, when  $OD_f$  is low,  $\beta$  is the same for all  $OD_f$  of a single spectrum and, hence, all wavelengths. This was observed earlier [16,26], but is not consistent with other observations [e.g., 8,12]. Explanations for a non-linear behavior of  $\beta$  with  $OD$  are particle shelf-shading effects and scattering problems in the determination of the real  $a_p$  in suspension [26]. Both are considered not to be typically relevant in the methodologies applied here (PSICAM and low  $OD_f$ ). And indeed, any non-linear behavior of  $\beta$  over  $OD_f$  of a single sample in the presented data set can be explained by a measurement error, most often due to an optically effective contamination of the CDOM filtrate in very clear waters. A linear behavior of  $\beta$  simplifies a quality control of the final  $a_p$  results, when  $\beta$  needs to be determined with an independent measurement. Any non-linearity between QFT and suspension measurements then indicates measurement errors. Nevertheless, without an independent  $a_p$  measurement of particles in suspension, the accuracy (in terms of absolute values) of the QFT technique is limited. Fortunately, if  $\beta$  is constant over  $OD_f$  for a single sample filter, several instruments and method exists that can measure  $a_p$  in suspension (e.g., AC-S WetLabs). These methods will partly have scattering error problems, but only one correct  $a_p$  values (at one wavelength) is needed to determine  $\beta$ .

The QFT-ICAM has the potential to simplify QFT-measurements, by allowing fast and easily repeatable measurements without a necessary sample preservation step. It will facilitate accurate determinations of particulate absorption coefficients in natural waters for the visible to short infrared spectral region. Future work will be dedicated to particulate light absorption in the ultraviolet, correction of effects induced by fluorescence from phycobiliproteins, and the causes for variation of the path length amplification in the filter.

## Acknowledgments

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