Proc. Estonian Acad. Sci. Biol. Ecol., 2003, **52**, 4, 423–436
https://doi.org/10.3176/biol.ecol.2003.4.05

Influence of light intensity on *in vivo* fluorescence characteristics of cyanobacteria

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Received 14 November 2002, in revised form 30 May 2003

Abstract. A laboratory experiment was carried out to estimate the extent of changes in fluorescence characteristics of three oscillatorian strains (*Planktothrix mougeotii*, *P. prolifica*, and *Tychonema bourrellyi*) grown in batch culture at two light intensities (9 and 39 μE m^{-2} s^{-1}). The following set of measurements was performed in the late log-phase of the growth of the cultures: recording of *in vivo* fluorescence spectra at 680 nm by excitation in the range 400–660 nm, determination of chlorophyll *a* and phycobilin concentrations, cells and trichome numbers, mean cellular volume, and dry weight of the culture. In *T. bourrellyi* and *P. prolifica* the lower light intensity stimulated the synthesis of phycoerythrin, and as a consequence the fluorescence response. Negligible changes in the pigment composition and shape of spectra of the *P. mougeotii* strain were observed. Differences in quantitative fluorescence characteristics normalized to phytoplankton biomass equivalents for cyanobacterial cultures grown at two light intensities are presented.

Key words: *in vivo* fluorescence, cyanobacterial phycobilins, light adaptation, monitoring of cyanobacterial blooms.

INTRODUCTION

Detection and observation of cyanobacterial blooms have become very topical all around the world. As cyanobacteria contain specific chromoproteins called phycobiliproteins, which have characteristic spectral structures at certain fluorescence excitation/emission co-ordinates (Rüdiger, 1980; Hilton et al., 1989a, b; Schubert et al., 1989; Kaitala et al., 1994; Poryvkina et al., 1994; Wood et al., 1998, 1999), they can be easily detected *in vivo* by measuring fluorescence spectra. All cyanobacteria contain at least three classes of blue phycobiliproteins: phycocyanin (PC), allophycocyanin (APC), and allophycocyanin B. A number of strains synthesize in addition red phycoerythrin (PE) or less commonly purplish phycoerythrocyanin (Chapman, 1973; Rüdiger, 1980; Carr & Wyman, 1986). Low chloro-

phyll (Chl) *a* emission while excited in its own absorption band at 430–440 nm is another peculiarity of fluorescence characteristics of cyanobacteria (Fig. 1). Quantitative (not functional) minority of the green pigment in these microorganisms' photosystem II, whose fluorescence dominates at normal temperature, is the reason of the phenomenon (Johnsen & Sakshaug, 1996). Prochlorophytes, or 'the other cyanobacteria' (Matthijs et al., 1994), form an exceptional group of blue-greens as they do not contain a detectable amount of phycobilins (PhBs) (Pinevich et al., 1999). Their fluorescence spectra are similar to those of green algae having maxima in the Chl *a* and *b* absorption range (Fig. 1).

An important step in applying *in vivo* fluorescence excitation spectra to water ecosystem analysis is moving from measurements of relative concentration of phytoplankton pigments to absolute values. To realize this step, the calibration dependences of fluorescence intensities on the pigment concentrations must be determined. Chl *a* is the only photopigment routinely determined in marine and freshwater samples.

Just a few attempts have been made to use the PhB fluorescence signal as an indicator of the abundance of the PhB-containing algal groups in natural water (Cowles et al., 1993; Lee et al., 1994; Poryvkina et al., 1994; Seppälä & Balode, 1998; Babichenko et al., 1999). Such rare use of *in vivo* fluorometry in the detection of cyanobacteria could have the following reasons:

1. The lack of a simple and reliable field technique to estimate PhBs' concentration hampers routine quantification of the PhBs' fluorescence signal in the processing of natural phytoplankton samples (Stewart & Farmer, 1984;

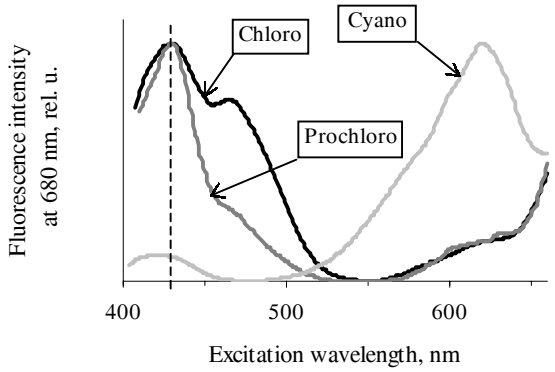

Fig. 1. Examples of *in vivo* fluorescence excitation spectra of algal and cyanobacterial monocultures from the culture collection of the Norwegian Institute for Water Research. Spectra are normalized to a maximum intensity in the spectrum. The dashed line marks an approximate excitation wavelength used widely in single-fixed-wavelength fluorometers for the determination of chlorophyll *a* fluorescence as a measure of this pigment concentration. Chloro = Chlorophyceae (*Pseudokirchneriella subcapitata*, strain CHL 1); Cyano = Cyanophyceae (*Pseudanabaena limnetica*, strain NIVA-CYA 276/6); Prochloro = Prochlorophyceae (*Prochlorothrix scandica*, strain 5/89).

Wyman, 1992). The main problem in the quantification of PhBs is to disrupt effectively a variety of cyanobacterial cells and achieve a complete extraction of the pigments as the cell walls of some blue-greens are extremely resistant to chemical and physical treatments (Rowan, 1989). On the other hand, care has to be taken not to overtreat samples; besides, they have to be handled in cold and dark as high temperature and intense light could degrade PhBs in extract (Moreth & Yentsch, 1970; Downes & Hall, 1998).

2. The quantity and quality of light can have a major effect on blue-greens, changing the pigment content, and as a consequence the fluorescence response (Hilton et al., 1989b). Alteration of PhB composition due to changes in light quality is well documented (Bennett & Bogorad, 1973; Bogorad 1975; Tandeau de Marsac, 1977). The phenomenon called complementary chromatic adaptation has not been observed in all cyanobacteria. Among those cyanobacteria that do not adapt chromatically to light quality picoplanktonic species have been reported (Carr & Wyman, 1986). Results of some studies suggest that the influence of light quantity on the pigment composition of cyanobacteria is also species-specific (Bryant, 1982; Carr & Wyman, 1986).

Although the response of pigment composition and concentration in cyanobacteria to quantitative and qualitative changes in light conditions has been investigated rather thoroughly, less is known of changes and the range of variations of *in vivo* fluorescence characteristics of blue-greens under different light conditions. The aim of the experiment reported here was to study the influence of light intensity on the quantitative and qualitative fluorescence characteristics of cyanobacteria. Measurements of *in vivo* fluorescence, pigment content, and biomass represented in cell and trichome numbers, cellular volume (on its basis calculation of cell carbon content is possible), and dry weight were carried out on three strains of cyanobacteria – *Planktothrix mougeotii*, *P. prolifica*, and *Tychonema bourrellyi* – to estimate the extent of variations of concentration-specific fluorescence characteristics under different light conditions. These potentially toxin-producing oscillatorians chosen for the experiment form dense blooms in eutrophic waters (Watanabe, 1979; Skulberg & Skulberg, 1985; Laugaste, 1986; Skulberg, 1999). The results obtained in the laboratory experiment can serve to provide a correct interpretation and quantitative transformation of fluorescence data registered during cyanobacterial blooms.

MATERIAL AND METHODS

Strains and growth conditions

Cyanobacterial strains from the algae culture collection of the Norwegian Institute for Water Research (NIVA) were used in the experiment. Detailed information on the strain isolated is given in Table 1. Precultures were grown in modified culture medium Z8 (Skulberg, 1990) at light intensity 6 $\mu E\ m^{-2}\ s^{-1}$ during 3 weeks. Fluorescent lamps (Philips TL 65W/33) were used for illumination. Cyanobacteria were cultivated on a shaking table at 18 °C.

Table 1. Information on the strains (isolated by R. Skulberg)

Species	Strain designation	Geographical origin	Year
Planktothrix mougeotii	NIVA-CYA 278	Lake Steinsfjorden, Buskerud, Norway	1989
Planktothrix prolifica	NIVA-CYA 98	Lake Steinsfjorden, Buskerud, Norway	1982
Tychonema bourrellyi	NIVA-CYA 33/1	Lake Mjøsa, Akershus, Norway	1976

For the experiment, an inoculate (20 mL) was added to 1 L of the above medium solution. The cultures were maintained under two light intensities: at 9 (D culture) and 39 μE m^{-2} s^{-1} (L culture). The samples for analyses were collected on the late log-phase of the growth of the cultures estimated by fluorescence intensity and Chl *a* concentration.

Fluorescence measurements

The *in vivo* excitation spectra of algae in the range 400–660 nm at the emission wavelength 680 nm were measured in a standard quartz cuvette with a spectrofluorimeter FluoImager 32B (Skalar). The spectral maxima measured for cyanobacteria reflect mainly the intensity of light emitted from Chl *a* by excitation of the PhBs; the corresponding maxima in the spectra are referred to as "Chl *a* fluorescence (at excitation around 430 nm), PE fluorescence (at excitation around 550 nm), and PC fluorescence (at excitation around 620 nm)". Culture samples with very high densities were diluted with culture medium before analysis. As the aim of the experiment was to analyse the relative changes in spectral features, no fluorescence spectral quantum correction was performed. Pigment-specific fluorescence was determined by normalizing the fluorescence intensity to the respective pigment concentration.

Pigment analysis

For the analysis of the amount of pigments (C_{pigment}) algal cells were collected by filtration on a glass fibre filter (type GF/F, Whatman) under suction pressure. Chl *a* concentration was measured in methanol extracts previously clarified by centrifugation at 49 500 *g* (outer row) for 1 h at 4 °C. *In vitro* absorption spectra were recorded with a UV-VIS scanning spectrometer Lambda 40 (Perkin Elmer). Absorptivity of 78.0 L g^{-1} cm^{-1} given by Talling & Driver (cited in Arvola, 1981) was used for the calculation of Chl *a*. For the determination of PhBs' concentrations cells were resuspended in 50 mM phosphate buffer (pH 6.75) and disrupted by sonication (VirSonic 300, Virtis Company) with a sonicator output of 60 W. The sonicator was run for 1 min by 20 s cycles with a 20 s break interval between the

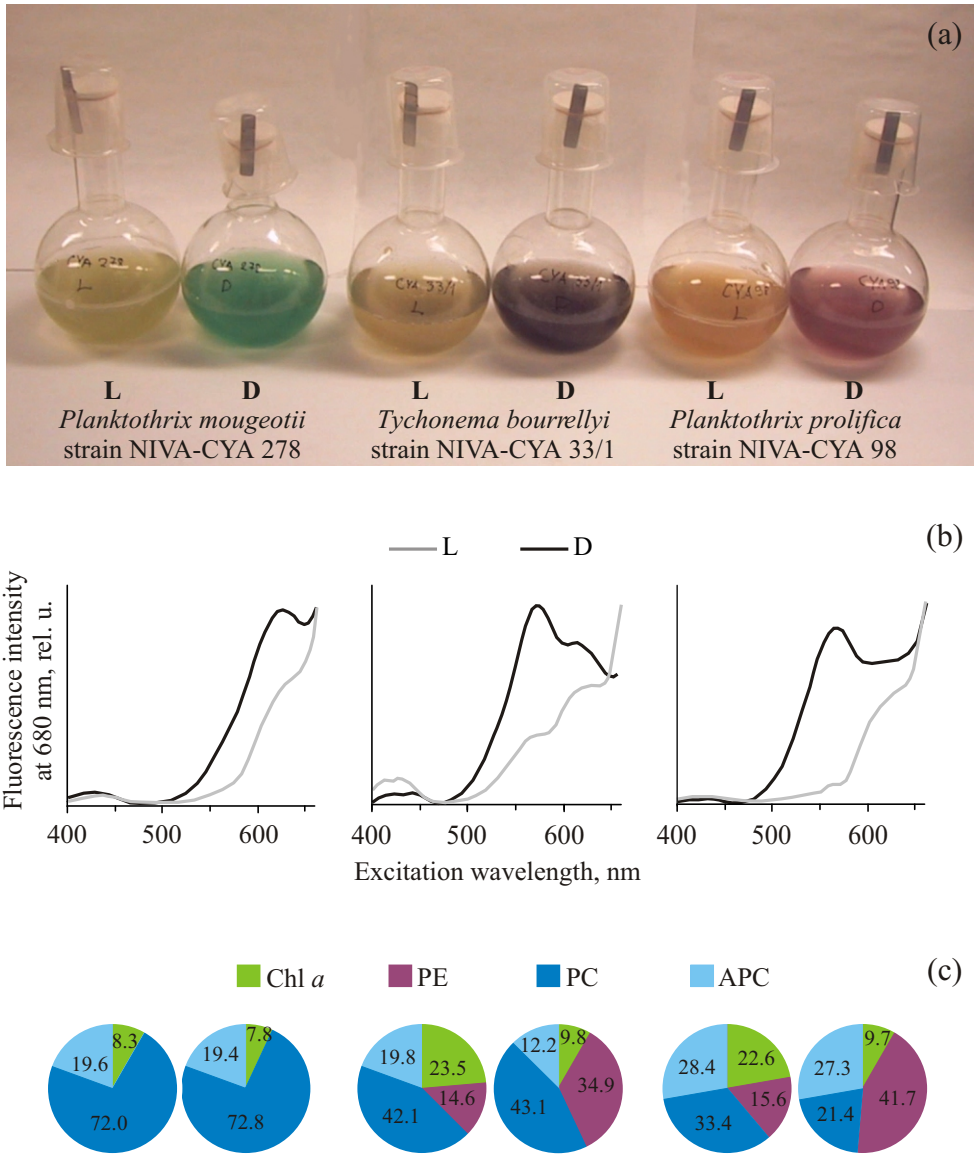

Fig. 2. Experiment on three cyanobacterial monocultures cultivated at two light intensities (39 and 9 ì E m^{-2} s^{-1}, in the figures L and D cultures, respectively). Differences in coloration (a), shape of *in vivo* fluorescence excitation spectra (b), and pigment content, % (c). Figures are placed under the corresponding culture in the photo. The photo was taken after 13 days of cultivation at different light illumination. Spectra and pigment concentration were obtained for all dark cultures on the 18th, for the L culture of NIVA-CYA 278 on the 11th, for the L culture of NIVA-CYA 33/1 on the 12th, and for the L culture of NIVA-CYA 98 on the 15th day after the start of the experiment. Fluorescence excitation spectra are scaled to a maximum intensity. The fluorescence maxima of excitation spectra correspond to the direct excitation of Chl *a* (in the violet spectral band) and its excitation through accessory pigments (phycoerythrin in yellow, phycocyanin in orange, and allophycocyanin in the red spectral band). Concentrations of cyanobacterial carotenoids, which serve mainly a photoprotective function, were excluded. Chl *a* = chlorophyll *a*; PE = phycoerythrin; PC = phycocyanin; APC = allophycocyanin.

runs to avoid heating up the extract solution. During all manipulations the tubes with extracts were kept in an ice bath. Before spectrophotometrical readings the extracts were clarified by centrifugation (at 51 000 *g* 4 h at 4 °C). The concentrations of PhB were calculated according to Bennett & Bogorad (1973).

Dry weight determination

The aliquots of cultures were filtered through a previously weighed glass fibre filter (type GF/C, Whatman). The filters were placed in a drying oven four 2 h at 104 °C. After that filters were reweighed and the dry weight (*DW*) of cyanobacterial cells in the aliquot was calculated.

Microscopic analysis

Cyanobacterial cells were preserved with Lugol solution. Cells and trichome (*tr*) numbers were counted by a light microscope. The average volume of the cyanobacterial cells (V_{cell}) was calculated using the stereometrical formula given by Edler (1979).

RESULTS

The experiment revealed marked differences in the coloration and fluorescence properties of cyanobacterial strains grown at the lower light intensity (Fig. 2a, b). The contrast in the pigmentation of cultures grown at the higher light was not so strong; however, an increased absorbance of light in the yellow spectral region was a distinctive pattern in the fluorescence excitation spectrum for the L culture of *T. bourrellyi*.

The spectra clearly indicate that *P. mougeotii* contained C-PC (excitation maximum occurred at 624 nm) and its PhB composition did not change due to light conditions. *T. bourrellyi* had both types of cyanobacterial PhB: C-PC (maximum at 615 nm) and C-PE (peak at 566 nm), but the relative intensity of PE fluorescence of this strain cultivated at the higher light was notably lower. The intensity of PE fluorescence (maximum at 569 nm) of the *P. prolifica* strain grown at the higher light illumination was very low and substituted for the excitation through PC. Analysis of the pigment concentration (Fig. 2c) showed also an increased portion of PE content in the D cultures of *T. bourrellyi* and *P. prolifica*. The red pigment in *P. prolifica* differed in its absorption properties from PE of *T. bourrellyi* (Fig. 3).

Changes caused by different light in cell and trichome level are given in Table 2. Higher number of cells and longer trichomes compared with L cultures were characteristic features for all D cultures. In the L cultures of *T. bourrellyi* and *P. prolifica* cells were larger than in the respective D cultures, contrary to the cultures of *P. mougeotii*.

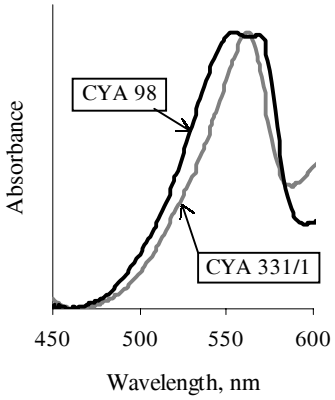

Fig. 3. Differences in absorption properties of phycoerythrins in phosphate buffer (pH 6.75) isolated from cyanobacterial strains *Tychonema bourrellyi* (CYA 33/1) and *Planktothrix prolifica* (CYA 98) grown at 9 μE m^{-2} s^{-1}. Peaks at 554, 568 (CYA 33/1), and 562 nm (CYA 98).

The main changes in quantitative fluorescence characteristics of the oscillatorian strains studied are summarized in Table 3. These data show that light intensity had a very low influence on the pigment-specific fluorescence of the *P. mougeotii* strain, but the pigments fluorescence calculated per cell, cellular volume, and trichome for the D culture was somewhat higher than in the L culture.

Cultivation of *T. bourrellyi* at the higher light illumination caused a significant drop in pigments-specific fluorescence. Miscellaneous behaviour of pigments fluorescence per biomass equivalents was observed: the lower light caused a decrease in the values for Chl *a* fluorescence. The corresponding parameters of PhBs were higher for the D culture, with the exception of PC fluorescence determined per cell. It should be noted that differences between D and L cultures of PE fluorescence per biomass equivalents were approximately 2-fold higher than the corresponding values for PC.

Table 2. Changes in cell densities, trichome numbers, mean cellular volume, and mean trichome length of three cyanobacterial cultures grown at 39 (L) and 9 μE m^{-2} s^{-1} (D)

Characteristic	*Planktothrix mougeotii*			*Tychonema bourrellyi*			*Planktothrix prolifica*		
	L	D	D/L	L	D	D/L	L	D	D/L
Cell numbers μL^{-1} (10^3)	3.8	6.8	1.8	2.0	10.1	5.1	4.1	9.0	2.2
Mean cell volume, μm^3	49.1	61.3	1.3	110.4	54.5	0.5	95.8	69.6	0.7
Trichome numbers μL^{-1}	40.0	33.3	0.8	65.2	112.2	1.7	143.0	19.6	0.1
Mean trichome length, mm	0.24	0.64	2.7	0.08	0.25	3.1	0.09	1.05	11.7

Table 3. Differences in the concentration-specific fluorescence characteristics and fluorescence ratios for cyanobacterial cultures grown at 39 (L) and 9 μE m^{-2} s^{-1} (D). Pigment and dry matter concentration – mg L^{-1}; cell and trichome concentration – number per L; cellular volume – μm^3

Characteristic	*Planktothrix mougeotii*			*Tychonema bourellyi*			*Planktothrix prolifica*		
	L	D	D/L	L	D	D/L	L	D	D/L
Pigment-specific fluorescence									
F(exChl *a*)/Chl *a*	222.1	194.1	0.9	502.1	108.7	0.2	118.1	46.9	0.4
F(exPE)/PE	0	0	–	2292.3	571.2	0.3	871.5	493.5	0.6
F(exPC)/PC	431.5	394.1	0.9	1390.0	372.1	0.3	2680.1	776.8	0.3
Fluorescence per cell (10^{-6})									
F(exChl *a*)/cell	0.04	0.07	1.6	0.15	0.04	0.3	0.02	0.04	2.1
F(exPE)/cell	0	0	–	0.42	0.72	1.7	0.10	1.83	18.5
F(exPC)/cell	0.71	1.30	1.8	0.73	0.58	0.8	0.65	1.48	2.3
Fluorescence per trichome (10^{-6})									
F(exChl *a*)/*tr*	4.0	14.1	3.5	4.5	3.5	0.8	0.6	18.6	33.3
F(exPE)/*tr*	0	0	–	12.7	64.7	5.1	2.8	844.8	297.0
F(exPC)/*tr*	67.2	266.2	4.0	22.2	52.1	2.4	18.7	681.2	36.4
Fluorescence per cellular volume, (10^{-9})									
F(exChl *a*)/V_{cell}	0.9	1.1	1.3	1.3	0.7	0.5	0.2	0.6	2.9
F(exPE)/V_{cell}	0	0	–	3.8	13.2	3.5	1.0	26.3	25.4
F(exPC)/V_{cell}	14.5	21.5	1.5	6.6	10.6	1.6	6.8	21.2	3.1
Fluorescence per dry weight									
F(exChl *a*)/*DW*	n.e.	3.5	–	2.2	1.9	0.9	0.5	2.2	4.4
F(exPE)/*DW*	n.e.	0.0	–	6.2	35.3	5.7	2.6	101.2	38.9
F(exPC)/*DW*	n.e.	66.9	–	10.8	28.4	2.6	17.1	81.6	4.8
Ratio									
F(exPE)/*F*(exChl *a*)	0	0	–	2.8	18.6	6.6	5.1	45.4	8.9
F(exPC)/*F*(exChl *a*)	16.8	18.8	1.1	5.0	15.0	3.0	33.5	36.6	1.1
F(exPE)/*F*(exPC)	0	0	–	0.57	1.24	2.2	0.15	1.24	8.2
Adaptation ratio, *R**	–1.000	–1.000	–	–0.271	0.108	–	–0.736	0.107	–

* $R = \dfrac{F(\text{exPE}) - F(\text{exPC})}{F(\text{exPE}) + F(\text{exPC})}$ (Hilton et al., 1989b)

ex = excitation

n.e. = not estimated

In the D culture of *P. prolifica* the pigment-specific fluorescence was also lower than in the L culture. All fluorescence calculations per biomass varied in a similar way: the light intensity of 9 μE m^{-2} led to an increase in the values, while changes for the PE values (D/L) were 8–9-fold as high as the differences in the corresponding values for Chl *a* and PC.

DISCUSSION

The influence of light intensity of 9 μE m^{-2} s^{-1} on the pigmentation and fluorescence spectra of two cyanobacterial strains – *T. bourrellyi* and *P. prolifica* – was similar to the effect of green light stimulating the synthesis of PE contrary to 39 μE m^{-2} s^{-1}, which like red light, reduced its synthesis in cells. On the whole, variation of fluorescence excitation spectra can be described using the chromatic adaptation ratio, *R* (Table 3), suggested by Hilton et al. (1989b). Theoretically *R* will vary from +1 to −1, respectively, when Chl *a* fluorescence at 680 nm is stimulated by PE excitation or by PC only. Comparison of *R* values for *T. bourrellyi* grown at intensities 9 and 39 μE m^{-2} s^{-1} in the experiment (Table 3) with *R* values calculated by Hilton et al. (1989b) for *Oscillatoria bourrellyi* (= *T. bourrellyi*) cultivated at 21 and 388 μE m^{-2} s^{-1} (*R* values 0.513 and 0.233, respectively) indicates that changes in fluorescence excitation spectra caused by different illumination are strain-specific and depend also on the photoacclimation status of cells.

As some blue-greens can produce PE, they are able to develop stable populations at depths where the light level is even less than 1% of surface illumination (Skulberg, 1999). However, cyanobacteria can be exposed to very high irradiances when they float up, or when they are passively carried up to the surface by vertical mixing processes in the water cplanktolumn. According to Demeters et al. (1991), physiological responses of phytoplankton to light fluctuations involve several mechanisms, observed as changes in (1) the fluorescence yield, (2) the size and density of photosynthetic units, (3) the composition and distribution of pigments, and (4) the chemical composition and activity of photosynthetic enzymes. These mechanisms respond at different rates and may depend on such factors as previous light and nutrient history of phytoplankton cells, the range of light intensity, light shift, and length of the exposition time (Prézelin & Matlick, 1983; Prézelin et al., 1986).

In the experiment the light conditions varied in a small range; with increasing culture density the actual level of light intensity certainly dropped even lower than the ambient illumination, especially for D cultures. A larger variation of light intensity can have a stronger effect on the fluorescence characteristics of cyanobacteria.

The higher illumination led to an augmentation of pigment-specific fluorescence indicating a decline in photosynthetic activity in the L cultures; the coloration and shortened trichomes of L cultures also point to unfavourable light conditions for these cyanobacteria strains. The L culture of *P. mougeotii* reacted to the higher light most drastically collapsing suddenly on the 11th day of the experiment. The collapse of the culture was determined from very low fluorescence intensity and extracted Chl *a* concentration; visually no changes in the culture colour were observed. However, according to Vila & Abella (2001), who investigated light-harvesting adaptation of planktonic phototrophic microorganisms in natural conditions, cyanobacteria rich in PC occupy an upper position in the water column compared to PE-containing cyanobacteria, which are dominant in deep metalimnia

(deeper than 8–10 m). Hence, *P. mougeotii* should endure higher light intensities better than red cyanobacteria.

Depending on the type and aim of investigations, different expressions of the fluorescence signal (F) in algal biomass units can be used. The experiment showed that differences in the concentration-specific fluorescence for D and L cultures (D/L) increased in the following order (for comparison corresponding values for Chl *a* fluorescence are also given):

P. mougeotii

Chl *a,* PC: $F/C_{\text{pigment}} \rightarrow F/V_{\text{cell}} \rightarrow F/\text{cell} \rightarrow F/tr$

T. bourrellyi

Chl *a*: $F/DW \rightarrow F/tr \rightarrow F/V_{\text{cell}} \rightarrow F/\text{cell} \rightarrow F/C_{\text{pigment}}$
PE: $F/\text{cell} \rightarrow F/C_{\text{pigment}} \rightarrow F/V_{\text{cell}} \rightarrow F/tr \rightarrow F/DW$
PC: $F/\text{cell} \rightarrow F/V_{\text{cell}} \rightarrow F/tr \rightarrow F/DW \rightarrow F/C_{\text{pigment}}$

P. prolifica

Chl *a*: $F/\text{cell} \rightarrow F/V_{\text{cell}} \rightarrow F/C_{\text{pigment}} \rightarrow F/DW \rightarrow F/tr$
PE: $F/C_{\text{pigment}} \rightarrow F/\text{cell} \rightarrow F/V_{\text{cell}} \rightarrow F/DW \rightarrow F/tr$
PC: $F/\text{cell} \rightarrow F/V_{\text{cell}} \rightarrow F/C_{\text{pigment}} \rightarrow F/DW \rightarrow F/tr$

This means that by converting the fluorescence signal from the cyanobacterial cells with different light adaptation status to biomass units, dispersion of data could be minimal for biomass equivalents represented at the beginning of the above-given series. Therefore, transformation of the fluorescence signal to the corresponding pigment concentration is widely used. However, high correlation between the fluorescence intensity of PhBs and the abundance of some cyanobacteria, expressed either by cell number or cellular volume, can also be achieved.

CONCLUSIONS

Information on the variability of the fluorescence of phycobilin pigments is important when excitation spectra of *in vivo* fluorescence are used in environmental studies. The illumination experiment on three strains of oscillatorians clearly demonstrated high variability and multi-directional changes of concentration-specific fluorescence characteristics in response to different light intensities. In this study changes in only phycoerythrin and phycocyanin were analysed. Registration of the Chl *a* fluorescence excitation spectrum further in the infrared region will allow recording the portion of Chl *a* fluorescence stimulated also by allophycocyanin. Considering this additional spectral part in analysis, it will be possible to distinguish spectrally more cyanobacterial (sub)groups, not only the PE and PC fluorescence group. As a result, an enhanced detection and identification of dominant species/groups of cyanobacterial blooms in automatic mode could be achieved.

ACKNOWLEDGEMENTS

This experiment and a part of the analyses were carried out at the Norwegian Institute for Water Research (NIVA). I am very grateful to the whole staff of the institute for hospitality and the opportunity to use laboratory facilities. My special thanks are due to Dr. Olav Skulberg and Torsten Källqvist for valuable theoretical and practical advice. I gratefully acknowledge Randi Skulberg for help in handling algal cultures. I also wish to thank my colleagues from the Institute of Ecology at Tallinn Pedagogical University Elga Rull for microscopic analysis of samples and Dr. Kai Künnis for valuable comments during preparation of the manuscript. I am very grateful to Prof. Jiří Komárek (Institute of Botany, Academy of Sciences of Czeck Republic) for encouragement and helpful discussion. The suggestions of two anonymous reviewers improved the paper considerably. This study was supported by the Research Council of Norway (grant IS-OE9 77/99) and the Estonian Science Foundation (grant 4714).

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Valguse intensiivsuse mõju tsüanobakterite *in vivo* fluorestsentsi karakteristikutele

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Viimastel aastakümnetel on kogu maailmas muutunud aktuaalseks sinivetikate "õitsengute" avastamine, nende leviku ulatuse kindlakstegemine ja biomassi määramine. Sinivetikate ehk tsüanobakterite olemasolu veeproovides on võimalik tuvastada spektraalse fluorestsentsi meetodi abil *in vivo*. Fluorestsentsi andmete interpreteerimisel peab arvestama, et valgustingimused võivad mõjutada sinivetikate pigmentkoostist ja järelikult ka spektraalkarakteristikuid. Fluorestsentsi karakteristikute muutuste ulatuse hindamiseks korraldati laboratoorne eksperiment kolme liigiga seltsist Oscillatoriales (*Planktothrix mougeotii*, *P. prolifica* ja *Tychonema bourrellyi*), mida kasvatati valguse intensiivsusel 9 ja 39 μE m^{-2} s^{-1}. Sinivetikate kultuuride hilises kiire kasvu faasis registreeriti spektraalses vahemikus 400–660 nm ergastatud pigmentide *in vivo* fluorestsents lainepikkusel 680 nm. Paralleelselt määrati klorofüll *a* ja fükobiliinide kontsentratsioon kultuuris, rakkude ja trihhoomide arvukus, keskmine rakumaht ning kuivaine kogus. Madalamal valgusintensiivsusel kasvatatud *T. bourrellyi* ja *P. prolifica* kultuurides suurenes fükoerütriini kontsentratsioon. Vastavad muutused olid jälgitavad ka fluorestsentsi ergastusspektrites. *Planktothrix mougeotii* kultuuride pigmentkoostist ja kvalitatiivseid spektraalkarakteristikuid mõjutas valguse intensiivsus vähe. Eksperiment näitas olulisi erinevusi valges ja hämaras kasvatatud sinivetikakultuuride fükobiliinide fluorestsentsi intensiivsuses sõltuvalt kasutatud biomassi ekvivalendist (pigmendi kontsentratsioon, vetikarakkude arvukus jne).