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Adaptation of algae to extreme environments

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Annotation

The acidophilic red alga *Cyanidium* sp. is one of the dominant mat-forming species in the highly acidic waters of Río Tinto, Spain. The culture of *Cyanidium* sp., isolated from a microbial mat sample collected at Río Tinto, was exposed to 9 different pH conditions in a gradient from 0.5 to 5 for 24 h and its physiological status evaluated by variable chlorophyll a fluorescence kinetics measurements. The optimum pH ranged from 1.5 to 2.5. A steep decrease of the photochemical activity was observed at pH below 1 even after 30 min of exposure. At pH above 2.5, the decline was more moderate and its negative effect on photochemistry was less severe. The red alga *Cyanidium* sp. is well-adapted to prevailing pH at its original locality at Río Tinto, i.e. pH of 1 to 3. The short-term survival in pH < 1.5 may be adaptation to rare exposures to such low pH in the field. The tolerance of pH above 3 could be caused by adaptation to the microenvironment of the inner parts of microbial mats in which *Cyanidium* sp. usually dominates and where higher pH could occur due to photosynthetic oxygen production.

Declaration [in Czech]

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ASTROBIOLOGY

Photochemical Performance of the Acidophilic Red Alga *Cyanidium* sp. in a pH Gradient

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Abstract The acidophilic red alga *Cyanidium* sp. is one of the dominant mat-forming species in the highly acidic waters of Río Tinto, Spain. The culture of *Cyanidium* sp., isolated from a microbial mat sample collected at Río Tinto, was exposed to 9 different pH conditions in a gradient from 0.5 to 5 for 24 h and its physiological status evaluated by variable chlorophyll *a* fluorescence kinetics measurements. Maximum quantum yield was determined after 30 min, 1 h, 2 h, 4 h, 6 h and 24 h of exposure after 15 min dark adaptation. The effect of pH on photochemical activity of *Cyanidium* sp. was observable as early as 30 min after exposure and the pattern remained stable or with only minor modifications for 24 h. The optimum pH ranged from 1.5 to 2.5. A steep decrease of the photochemical activity was observed at pH below 1 even after 30 min of exposure. Although the alga had tolerated the exposure to pH=1 for at least 6 h, longer (24 h) exposure resulted in reduction of the photochemical activity. At pH above 2.5, the decline was more moderate and its negative effect on photochemistry was less severe. According to the fluorescence measurements, the red alga *Cyanidium* sp. is well-adapted to prevailing pH at its original locality at Río Tinto, i.e. pH of 1 to 3. The short-term survival in pH<1.5 may be adaptation to rare exposures to such low pH in the field. The tolerance of pH above 3 could be caused by adaptation to the microenvironment of the inner parts of microbial mats in which *Cyanidium* sp. usually dominates and where higher pH could occur due to photosynthetic oxygen production.

Keywords Acidic environment · Algae · Chlorophyll fluorescence · Cyanidium · pH gradient

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Introduction

The study of life in extreme environments is essential for understanding how life established on the early Earth and for assessing the possibilities of life on other planetary bodies (Gómez et al. 2011). Such terrestrial environments could serve as terrestrial analogues of extraterrestrial conditions due to similarities in geology, mineralogy, chemistry of the system, temperature, salinity, or dryness (Gómez 2011). The highly acidic river Río Tinto, Spain, is considered as a geochemical analogue of Mars since its mineralogy resembles the mineralogy of the Meridiani Planum region on Mars (Fernández-Remolar et al. 2005; Squyres and Knoll 2005). Knowledge about the physiological diversity of microorganisms in such an analogue could be beneficial for the search of life beyond Earth in order to determine the limits of carbon-based life (Baross et al. 2007).

Oxygenic eukaryotic phototrophic microorganisms, algae, are one of the main primary producers in the very acidic waters of Río Tinto. Their biomass reaches more than 60 % of total biomass in the river (Aguilera et al. 2007c; López-Archilla et al. 2001); usually they are associated in microbial biofilms (Aguilera et al. 2007a, b). The variable chlorophyll fluorescence measurements of the Río Tinto phototrophic biofilm proved the relatively good physiological status of the biofilm (Gómez et al. 2011), however the biofilm consists of different algal species and thus, the resulting fluorescence parameters reflect an averaged response of all individuals in the biofilm. Values of the fluorescence parameters would be dependent on the biofilm-forming species composition and the response of individual species to the particular environmental conditions. In order to explain the collected data, knowledge about species composition and individual species response to various environmental factors is necessary, however these data on individual response of each mat forming species remains to be determined.

One of the dominant biofilm-forming species is the unicellular red alga *Cyanidium* sp., especially in the most acidic conditions (Aguilera et al. 2007b). Several studies on its ecophysiology proved that this microalga is well adapted to low pH and high temperature (Ascione et al. 1966; Doemel and Brock 1971; Enami 1978; Enami and Fukuda 1975, 1977a, b, 1978; Enami et al. 1975, 1978; Revsbech and Ward 1983; Wollman 1979). Surprisingly, studies using variable chlorophyll fluorescence measurements in *Cyanidium* sp. are rare (Diner 1979; Diner and Wollman 1979) and not focused on ecophysiological characteristics. This well-established physiological method (see e.g. Bohlar-Nordenkampf et al. 1989; Krause and Weis 1991; Kromkamp and Forster 2003; Maxwell and Johnson 2000) is becoming a useful tool for field studies in extreme environments (Gómez et al. 2011; Kvíderová 2010a; Kvíderová et al. 2011) and could provide new data on ecophysiological processes, either during in situ studies or laboratory experiments.

The aim of this study was to evaluate the short-term response of the photochemical processes of one of the dominant mat-forming species in Río Tinto, *Cyanidium* sp. (Rhodophyceae), to a pH gradient using the variable chlorophyll fluorescence approach and to compare these results with data from field measurement in Río Tinto (Gómez et al. 2011). These data would improve the explanation of field data obtained in the future during in situ fluorescence measurements in acidic environments. These data will also be included in a database focused on the taxonomic, biochemical and physiological diversity of extremophilic cyanobacteria and algae which is being built in the Centre for Phycology, Institute of Botany AS CR in Třeboň, Czech Republic.

Material and Methods

Algal Cultivation and pH Treatment

The culture of the red alga *Cyanidium* sp. was isolated from a microbial mat collected at Río Tinto, Spain (see Gómez et al. 2011 for collection procedure details). The strain is kept in the *Cyanidium* medium of pH=2.5 (Andersen 2005) at 30 °C and 50 μmol m^{-2} s^{-1} at the Institute of Botany in Třeboň, Czech Republic.

Before exposure, the cell suspension was washed twice with 0.5 % (w/v) KCl by centrifugation at 1,000 g/25 °C/15 min. The dense suspension was inoculated into fresh *Cyanidium* medium of pH ranging from 0.5 to 5. Nine pH exposures (pH of 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4 and 5) and control (original stock culture without washing) were used. Three replicates were used in each treatment. The cells were exposed to the actual pH and their photochemical performance evaluated after 30 min, 1 h, 2 h, 4 h, 6 h and 24 h after exposure.

During the experiment, the algae were kept at 25 °C and continuous irradiance of 50 μmol m^{-2} s^{-1} during exposure in plastic cuvettes suitable for fluorescence measurements and covered by the BF-400 breathable sealing film for tissue cultures (Axygen Bioscience, USA). The volume of the suspension in the cuvette was 1.2 ml.

Variable Chlorophyll *a* Fluorescence Measurement

Photochemical activity measurements were performed using a Dual Modulation Fluorometer (Photon Systems Instruments, Czech Republic). Q_A-reoxidation was measured according to Kvíderová (2010b). The samples were dark-adapted for 15 min. One single-turnover saturating pulse lasting 15 μs was applied after the start of the experiment. The maximum quantum yield (F_V/F_M) was calculated using the formula of Roháček and Barták (1999):

$$F_V/F_M = \frac{F_M - F_0}{F_M}$$

where F_0 is the minimum fluorescence in the dark and F_M is the maximum fluorescence after the saturation pulse. The fluorescence decay half-time was estimated from the decay kinetics during the first second of fluorescence quenching by fitting a triple exponentional decay equation (Kolber et al. 1988):

$$\Delta\Phi(t) = ae^{(-t/\tau)} + be^{(-t/\tau_2)} + ce^{(-t/\tau_3)}$$

where $\Delta\Phi(t)$ is the change of the fluorescence signal over time, t; a, b and c are parameters describing the amplitude; τ, τ_1 and τ_2 are rates of transport within photosystem 2. The fitting was performed using SigmaPlot 10.0 softwate (Systat Software, USA).

Statistics

Statistical significance was evaluated using Statistica 10.0 (StatSoft, USA). Homologous groups were distinguished by Tukey's Honest Significant Diifference (HSD) test for the significance level P=0.05. ANOVA was used to test the time changes in the F_V/F_M and a paired *t*-test was used to compare changes in the rate of transport over time. The results were considered significant for P<0.05.

Results

The F_V/F_M values indicated a relatively good physiological state of the cultures at optimum pH conditions and the control at the start of the experiment. The washing of the cultures by 0.5 % KCl, and hence short-term rapid pH changes, did not affect the photochemical performance of the cultures used for the exposure experiment, since there were no significant differences between the control and the culture exposed to pH 2.5 (Table 1).

The effects of pH on F_V/F_M were observable almost immediately (30 min) after exposure. The optimum pH, defined as the pH in which the mean change of the F_V/F_M was less than 5 % of the control value after 30 min of exposure (C_{30min}) during the experiment, ranged from 1.5 to 2.5. However, the response of *Cyanidium* sp. was different at low and high pH exposures. pH values lower than optimum were considered as "low pH" ranging from pH 0.5 to pH 1. In low pH, rapid changes in F_V/F_M occurred during the experiment, probably due to the destruction of cell membranes almost immediately at pH 0.5 or after 24 h of exposure at pH 1. pH values higher than optimum were considered as "high pH" ranging from pH 3 to pH 5. At high pH values in the range from 3 to 5, the decline of F_V/F_M was milder than in pH 0.5, probably due to better adaptation to higher pH (Table 1, Fig. 1).

As seen in Fig. 2, the F_V/F_M values at individual pH exposures remained stable for at least 24 h with the exception of pH 1. The cultivation conditions could not affect the fluorescence measurement results, since the F_V/F_M values of the control during the experiment ranged from 98.6±1.5 to 101.4±0.5 % of $C_{30\ min}$ (mean ± standard deviation, n=3 for each exposure duration), and no statistically significant differences were detected for exposure duration. In the optimum pH range, there was no significant time changes in F_V/F_M (one-way ANOVA, n=18, P>0.05 in all cases, Fig. 2), and the change of F_V/F_M did not fall under 11 % of the $C_{30\ min}$ proving thus good physiological status of *Cyanidium* sp.

A different response to low pH was observed at pH 0.5 and pH 1. At pH 0.5, a very steep drop of F_V/F_M by 32.5±6.5 % (mean ± standard deviation, n=3) was observed even after 30 min exposure duration and did not change significantly during the experiment. On the other hand, F_V/F_M remained ca 12 % lower than $C_{30\ min}$ during the first 6 h of exposure to pH 1, and a sudden decrease to by 24.1±2.1 % compared to $C_{30\ min}$ was detected after 24 h (one-way ANOVA; n=18; P=0.001; Fig. 2), probably due to temporal resistance to pH 1.

Contrary to the low pH, all high pH exposures had similar responses. The F_V/F_M decreased to ca 15–20 % of $C_{30\ min}$ in the first 30 min of the experiment. No statistically significant F_V/F_M changes occurred over time indicating either slow damage to the cells (decrease in F_V/F_M) or acclimatization (increase in the F_V/F_M) (one-way ANOVA, n=18, P>0.05 in all cases, Fig. 2).

Due to the similarity of the response to pH during the first six hours of exposure, only the fluorescence decay parameters data from 1 h and 24 h exposures are shown (Table 2, Fig. 3). The change of the decay curves at pH 1 (Fig. 3) in time indicated a faster decay in suboptimal conditions reflecting thus possible membrane damage in pH 0.5 and 1. However, this was not supported by decay curve parameters. Generally, the curve parameter values were less sensitive to the pH treatments than F_V/F_M and only minor changes were observed after short-term exposure at pH 0.5 in τ and τ_1 and no significant differences were observed after 24 h exposure, probably due to the larger variability in samples (Table 2). The parameter values measured after 1 and 24 h did not differ significantly in individual pH exposures (paired t-test, P>0.05 in all cases).

Table 1 Values of F_V/F_M (mean ± standard deviation; n=3 for each treatment) at individual pH exposures and exposure durations

Exposure duration / pH	30 min	1 h	2 h	4 h	6 h	24 h
0.5	0.286±0.038^{c}	0.287±0.016^{c}	0.313±0.032^{c}	0.308±0.019^{c}	0.283±0.041^{b}	0.293±0.018^{e}
1	0.365±0.006^{abc}	0.371±0.005^{abc}	0.381±0.015^{abc}	0.365±0.013^{bc}	0.369±0.013^{a}	0.320±0.004^{de}
1.5	0.412±0.034^{ab}	0.412±0.017^{a}	0.426±0.010^{a}	0.424±0.010^{a}	0.421±0.021^{a}	0.406±0.010^{ab}
2	0.398±0.027^{ab}	0.399±0.018^{ab}	0.403±0.018^{ab}	0.406±0.019^{ab}	0.412±0.027^{a}	0.398±0.023^{abc}
2.5	0.377±0.034^{abc}	0.394±0.017^{ab}	0.407±0.027^{ab}	0.419±0.010^{ab}	0.419±0.030^{a}	0.400±0.011^{abc}
3	0.354±0.038^{abc}	0.365±0.039^{abc}	0.381±0.027^{ab}	0.403±0.017^{ab}	0.389±0.013^{a}	0.376±0.009^{abc}
3.5	0.339±0.030^{abc}	0.349±0.036^{bc}	0.353±0.030^{bc}	0.380±0.023^{bc}	0.375±0.024^{a}	0.364±0.012^{bcd}
4	0.326±0.045^{bc}	0.331±0.032^{bc}	0.341±0.035^{bc}	0.355±0.046^{bc}	0.355±0.039^{ab}	0.353±0.017^{cd}
5	0.339±0.036^{abc}	0.341±0.030^{bc}	0.341±0.024^{bc}	0.347±0.032^{bc}	0.349±0.028^{ab}	0.355±0.028^{cd}
Control	0.423±0.016^{a}	0.426±0.015^{a}	0.428±0.016^{a}	0.429±0.018^{a}	0.424±0.018^{a}	0.417±0.016^{a}

The superscript letter(s) indicate(s) the homologous group distinguished by the Tukey's HSD test at a significance level of P=0.05; this analysis was performed for each exposure duration independently. The group indicated by "a" includes control

Discussion

So far, no study of ecophysiological characteristics of the acidophilic red alga *Cyanidium* sp. has been performed using measurements of variable chlorophyll *a* fluorescence under laboratory conditions. The fluorescence measurements in *Cyanidium* sp. isolated from the acidic Río Tinto showed that pH optimum for photochemical reactions ranged from 1.5 to 2.5. Lower pH led to a rapid and steep decrease of F_V/F_M indicating thus probably serious cell damage at pH 0.5 and 1. However, the medium decline of F_V/F_M and a stable response to high pH could be caused by adaptation to the biofilm microenvironment. The found photochemistry optimum was comparable to optima determined previously from oxygen evolution measurements (Enami and Fukuda 1975) or growth experiments (Doemel and Brock 1971; Gimmler 2001), and probably reflected adaptation/acclimatization to the prevailing conditions at the original locality in Río Tinto, i.e. annual mean pH~2.2 (López-Archilla et al. 2001).

Since pH values below ~1.5 usually did not occur in situ and were very rare and episodic (Aguilera et al. 2006, 2007c; Lopez-Archilla and Amils 1999; López-Archilla et al. 2001; Souza-Egipsy et al. 2008), there was only negligible selection pressure for adaptation/acclimatization to such low pH which could result in the limited resistance to pH below 1.5. Moreover, low pH tolerance could be significantly influenced by light conditions. In darkness, the resistance of the acidophilic red algae *Cyanidium caldarium* and *Galdieria sulphuriana* to low pH could last even several months (Gross 2000). However, photooxidative damage of membranes caused by high light (Ledford and Niyogi 2005) could contribute to increased sensitivity to low pH, most likely due to increased membrane permeability for protons (Gross 2000).

On the other hand, the better resistance/tolerance at pH≥3 could rather indicate adaptation to conditions in biomat interiors, since pH values higher than 3.05 were not observed in the waters of Río Tinto (López-Archilla et al. 2001). Due to intensive photosynthetic oxygen production in *Cyanidium* biofilm (Revsbech and Ward 1983), the internal pH could be significantly higher compared to the surroundings. Such increased internal pH accompanied

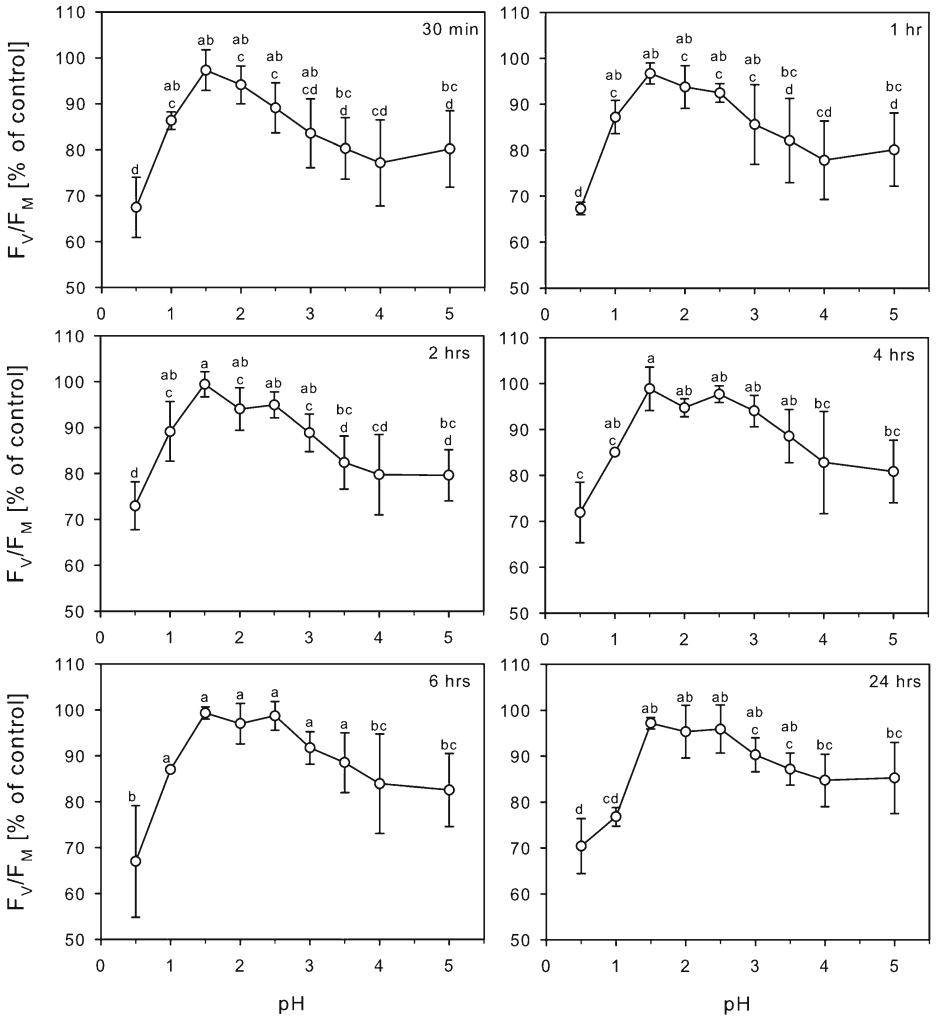

Fig. 1 Response of the maximum quantum yield (F_V/F_M; mean ± standard deviation; n=3 for each pH exposure) to the pH at different exposure durations, indicated by chart label. The F_V/F_M values were normalized to the F_V/F_M values of the control for each exposure and were expressed as percentage of control at a given exposure duration. The letter(s) at each chart datapoint indicates homologous groups defined by the Tukey HSD test at a significance level of P=0.05. The group indicated by "a" always includes control

by high oxygen concentration was already observed in the internal part of a microbial mat (Revsbech et al. 1983). Even in biofilm, the *Cyanidium* sp. could not probably survive at pH>5, since *Cyanidium caldarium* was found to occur in water or soil with a pH ranging from 3 to 5, but not above pH 5, in natural populations in Yellowstone National Park, U.S.A. (Doemel and Brock 1971).

The hypothesis of better tolerance at pH 3 in *Cyanidium caldarium* could be also supported by evolution of oxygen measurements. A second maximum of photochemical activity was observed at pH 7 under different light treatments; this could also indicate the tolerance of *Cyanidium caldarium* to pH 7 (Enami and Fukuda 1975). In experiments reported in this work, this second maximum of photochemical activity was not detected,

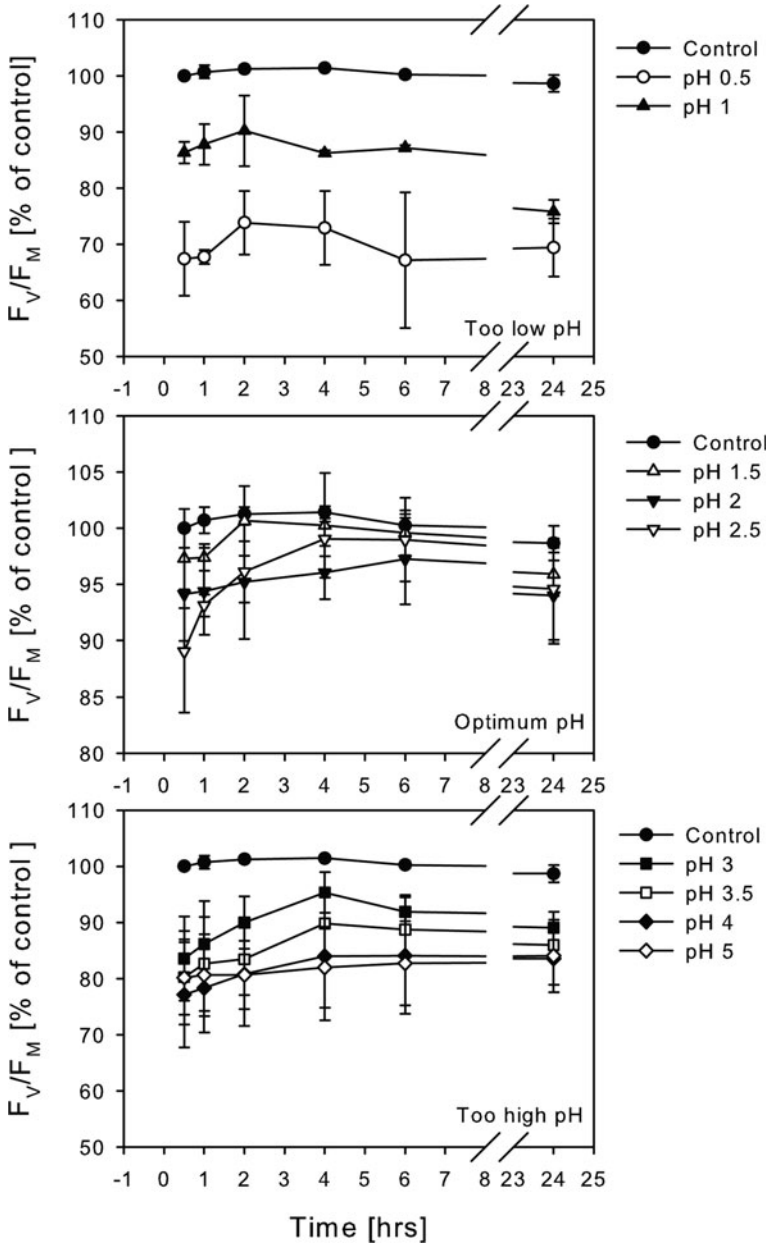

Fig. 2 The time course of the response of F_V/F_M at different pH exposures. The F_V/F_M values (mean ± standard deviation; n=3 for each pH exposure) were normalized to the F_V/F_M values of the control measured after 30 min after the start of the experiment. The individual charts are dividend into low pH (0.5–1), optimum pH (1.5–2.5) and high pH (3–5) groups

since neutral and basic pH exposures (above pH 7) were not possible due to the bad buffering capacity of the *Cyanidium* medium at pH>4, meaning that a pH 5 was the highest achievable in this medium.

Table 2 Parameters of the fluorescence decay curve (mean ± standard deviation; n=3 for each pH exposure) at individual exposures and exposure durations

Parameter pH	τ [μs]	τ_1 [ms]	τ_2 [ms]
1 h			
0.5	15.0±10.4[b]	0.04±0.01[b]	33.4±38.2[a]
1	25.5±18.0[ab]	0.31±0.45[ab]	30.3±24.9[a]
1.5	29.7±1.7[ab]	0.58±0.22[ab]	25.7±15.8[a]
2	43.9±13.9[a]	3.12±2.60[a]	252.3±239.2[a]
2.5	29.7±7.2[ab]	1.05±0.53[ab]	34.4±6.3[a]
3	34.9±6.7[ab]	0.70±0.57[ab]	72.2±58.7[a]
3.5	37.1±3.9[ab]	1.85±1.73[ab]	309.9±258.7[a]
4	31.3±4.5[ab]	0.43±0.25[ab]	18.9±11.8[a]
5	21.7±7.3[ab]	0.29±0.23[ab]	366.4±549.1[a]
Control	41.9±4.4[a]	1.81±0.23[ab]	95.8±33.0[a]
24 h			
0.5	8.1±17.6[a]	0.03±0.02[a]	178.4±308.9[a]
1	16.3±23.7[a]	0.04±0.00[a]	7.6±8.6[a]
1.5	34.2±2.4[a]	1.50±1.74[a]	95.9±11.2[a]
2	32.2±4.2[a]	1.22±0.70[a]	46.3±31.2[a]
2.5	27.9±16.0[a]	0.88±0.46[a]	24.3±18.2[a]
3	15.1±4.3[a]	0.04±0.00[a]	4.8±2.2[a]
3.5	35.4±308.4[a]	0.81±0.34[a]	133.8±58.5[a]
4	217.0±840.5[a]	6.84±11.80[a]	131.1±76.9[a]
5	605.7±0.3[a]	0.03±0.00[a]	92.2±28.6[a]
Control	39.4±17.6[a]	2.03±0.04[a]	221.7±73.6[a]

The superscript letter(s) indicate (s) homologous groups distinguished by the Tukey HSD test at a significance level of P= 0.05. The group indicated by "a" includes control

The time course response of F_V/F_M to different pH indicated a rapid (30 min) response of *Cyanidium* sp. to pH exposure, with the exception of pH 1 where transient resistance was observed. The response to pH exposure differed at low and high pH values. The rapid decrease of F_V/F_M at pH 0.5 likely indicated cell damage and failure of protective mechanisms. At pH 1, the alga was able to resist the stress conditions for at least for 6 h, but not longer than 24 h, probably due to some kind of transient protection. According to Gimmler (2001), the adaptation/protection mechanisms of acidohilic algae included proton export from a cell, reversed electrical potential on the membrane and modification of membrane architecture. At pH 0.5, membrane damage seemed to occur, since the photochemical reactions were not affected by acidity in intact cells only (Enami and Fukuda 1975, 1977a). The prolonged resistance to pH 1 could be probably caused either by a temporal increase in cell membrane impermeability for protons and/or a high internal proton buffer capacity (Gimmler 2001). The mild decline and stable response at pH≥3 may suggest adaptation reactions in which the above mentioned protective mechanisms participate, however the rates of equilibration of internal pH after external pH change are not known (Gimmler 2001).

Contrary to F_V/F_M, the values of the electron transport rates within photosystem II seemed to be only slightly modified by pH, with the exception of pH 0.5. The photosynthetic membrane, or at least photosystem II, probably remained intact for 24 h even in sub-optimal pH exposures. The faster fluorescence rate at pH 0.5 should indicate damage of internal

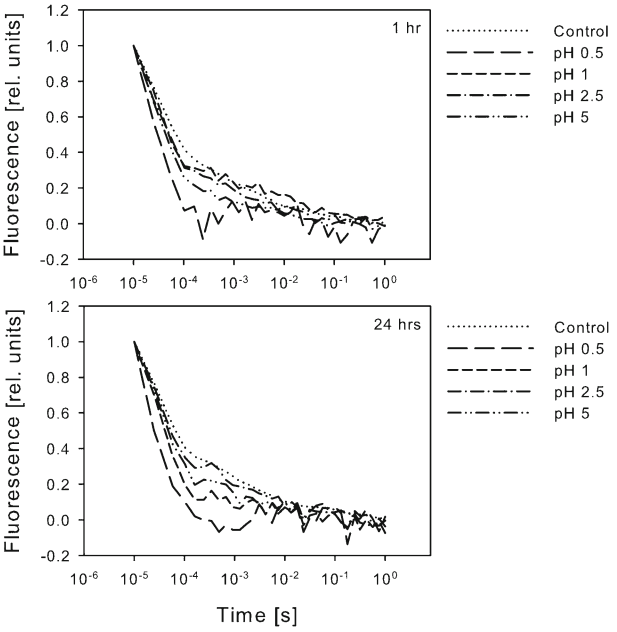

Fig. 3 Time courses of the fluorescence decay at 1 h and 24 h exposure duration indicated as chart label. The line corresponds to the mean (n=3 for each pH exposure and exposure duration) for individual treatments and time. The shown treatments represent low pH (pH 0.5 and 1), optimum pH (pH 2.5), high pH (pH 5) and a control

cellular structures. Since the decline of F_V/F_M generally indicates any stress conditions (Maxwell and Johnson 2000), damage of other cellular processes than membrane transport could be possible. Long-term (several days or weeks) experiments will be necessary to determine the damage and repair dynamics within photosystem II.

The optimum, i.e. maximum, values of F_V/F_M in this study were lower than those measured previously at Río Tinto (Gómez et al. 2011). The difference could be caused by different instrumentation and protocol selection as well as the species composition of the evaluated samples. Short (single-turnover) pulses lasting 15 μs were applied during Q_A-reoxidation protocol in the Dual Modulation Fluorometer device (see Trtílek et al. (1997) for detailed device description) in this study, but longer (multi-turnover) pulses lasting ca 1 s were applied during the field measurement (Gómez et al. 2011) using Quenching analysis protocol in the FluorCam device (see Nedbal et al. (2000) for detailed device description). Compared to single-turnover pulses, F_V/F_M determined by multi-turnover is approximately 10–15 % higher (Kromkamp and Forster 2003). Therefore, the fluorescence instrumentation (fluorometers vs. fluorescence imaging cameras) and measurement protocols (Q_A-reoxidation vs. Quenching analysis) must be considered when interpreting the data in future experiment proposals, as well as comparisons to previous studies.

The lower F_V/F_M values recorded in experiments reported in this work could also be caused by using pure *Cyadinium* sp. culture, omitting thus the strong signal from the filamentous green alga *Klebsormidium* sp. in the field samples (Gómez et al. 2011). The F_V/F_M values of *Klebsormidium* should be higher in the optimal conditions due to the different structure of the photosynthetic apparatus, as observed in other algal classes (Juneau and Harrison 2005).

Fluorescence measurements proved to be a valuable tool in ecophysiological characterization of extremophilic photosynthetic microorganisms (e.g. Gómez et al. 2011; Kvíderová 2010a, 2011) and could help to estimate their (eco)physiological diversity. At present, screening of the physiological diversity of Earth-life is one of the main issues being addressed by the astrobiological community. So far, only minor knowledge on metabolic and physiological diversity of a majority of microorganisms is available even from non-extreme environments; the study of the physiological diversity of extremophiles could lead to novel insights into adaptation/acclimatization mechanisms (Baross et al. 2007). For this purpose, databases on extremophilic and even non-extremophilic (micro)organisms, covering detailed taxonomic, geographical, biochemical and physiological data, should be established, since the present databases of microorganisms cover geographical and taxonomical diversity only (Floyd et al. 2005). The measurement of chlorophyll fluorescence, using a modulated signal with different excitation and detection wavelenghts, could be a useful tool for screening this photochemical variability of photoautotrophic life in extreme environments on Earth (e.g. Gómez et al. 2011; Kvíderová 2010a; Kvíderová et al. 2011), or even on exoplanets. In the later case, the star spectral class type must be considered (Kiang et al. 2007a, b).

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