

Photosynthetic activity of microphytobenthos measured *in situ* using chlorophyll fluorescence rapid light curves

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2 - *In situ* measurements

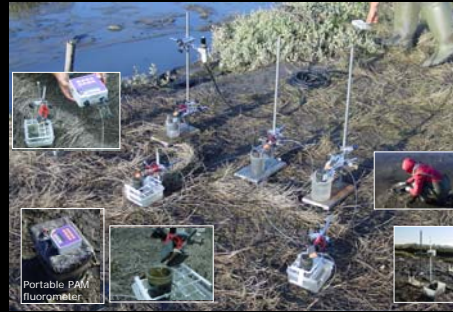
The photosynthetic activity of microphytobenthos (MPB) was studied using PAM fluorescence Rapid Light Curves (RLCs):

- 1 - The light response of RLCs and its relationship with non-photochemical quenching (NPQ) was characterised in MPB suspensions
- 2 - PAM fluorescence parameters (F_o , $\Delta F/F_m$, F_v/F_m) and RLCs were measured *in situ* with a frequency of 10-15 during low tides



Sampling site: intertidal muddy sediments on Canal de Ilhavo, Ria de Aveiro, Portugal; microalgae were collected during periods of low tide using the lens tissue technique

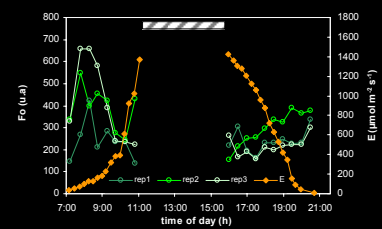
- RLCs were constructed by exposing the samples to 12 increasing irradiance (E) levels, from 21 to 1707 $\mu\text{mol m}^{-2} \text{s}^{-1}$; under each E, F_o and F_m were measured each 90 seconds until a steady-state in $\Delta F/F_m$ was reached; the relative transport electrons rate ($\text{ETR} = E \cdot \Delta F/F_m$) and the non-photochemical quenching coefficient were determined for each light level
- NPQ was determined as $\text{NPQ} = (F_{m,m} - F_m)/F_m$, where $F_{m,m}$ is the maximum F_m value measured.



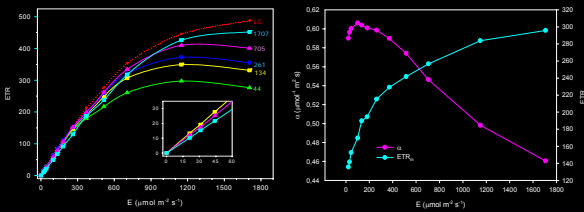
RLCs and fluorescence parameters were measured *in situ* using a portable PAM fluorometer (Junior-PAM, Gademán Instruments, Würzburg, Germany), using a 1.5 mm-diameter plastic fiberoptic

- 3 to 6 replicate measurements each sampling occasion
- Irradiance, salinity and temperature were also measured

Example of *in situ* time series of F_o , F_m and RLC parameters α and ETR_m .

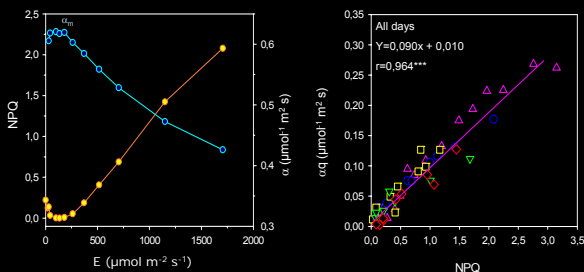
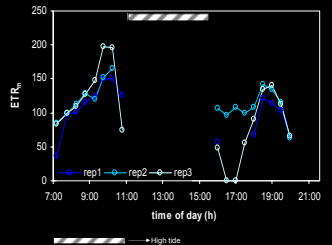
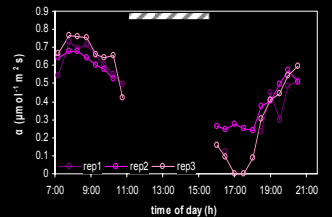


1 - Light response of RLCs and NPQ



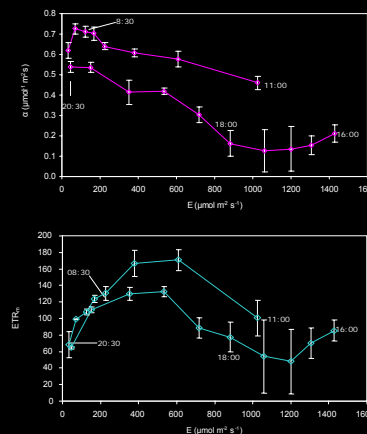
- Both light-limited (α) and light-saturated (ETR_m) parts of RLCs varied bi-phasically with light
- α increased under low light, and decreased significantly under high light
- ETR_m increased slightly under low light, and significantly under high light
- Steady-state light curves higher than RLCs

- F_o values increased until a maximum value reached in the middle of low tide and then decreased
- α increased under low light, followed by a decrease with irradiance above ca. 100 $\mu\text{mol m}^{-2} \text{s}^{-1}$
- ETR_m increased with irradiance until 600 $\mu\text{mol m}^{-2} \text{s}^{-1}$ after which, started to decrease
- Despite almost symmetrical variation of E during the two low tide periods, α and ETR_m displayed markedly different patterns
- Recovery of α and ETR_m during the second period of low tide was incomplete



- α and NPQ varied with light virtually symmetrical to each other: the initial increase in α under low light was paralleled by decrease in NPQ, and the α quenching under high light was followed by an increase in NPQ
- The increase of NPQ under high light was found to correlate significantly with the high light-induced quenching of α calculated by: $\alpha_q(E) = \alpha_m - \alpha(E)$
- The linear relationship between α_q and NPQ was verified on 5 days along a spring-neap tidal cycle

RLC parameters vs E



- To compare with the light response measured in the microalgal suspensions, α and ETR_m values measured *in situ* were plotted against E
- The light response of α during the first low tide period is similar to the expected from laboratory measurements, denoting the onset of NPQ processes
- The α light responses in the two low tide periods are markedly different: in the afternoon, recovery of α is incomplete, appearing to suffer from cumulative photoinhibitory effects of prolonged high light exposure
- The pattern of ETR_m light response is clearly different from the obtained in suspensions, showing a marked decrease under high light
- Cumulative effects are also present in the ETR_m light response measured in the second low tide period

Acknowledgements: This work was supported by Fundação para a Ciência e a Tecnologia, project PDCTM/MAR/15318/99.