
**Photosynthetic and respiratory gas exchange characteristics
of
Synechocystis PCC 6803 *ndhD*(1-4) mutants**

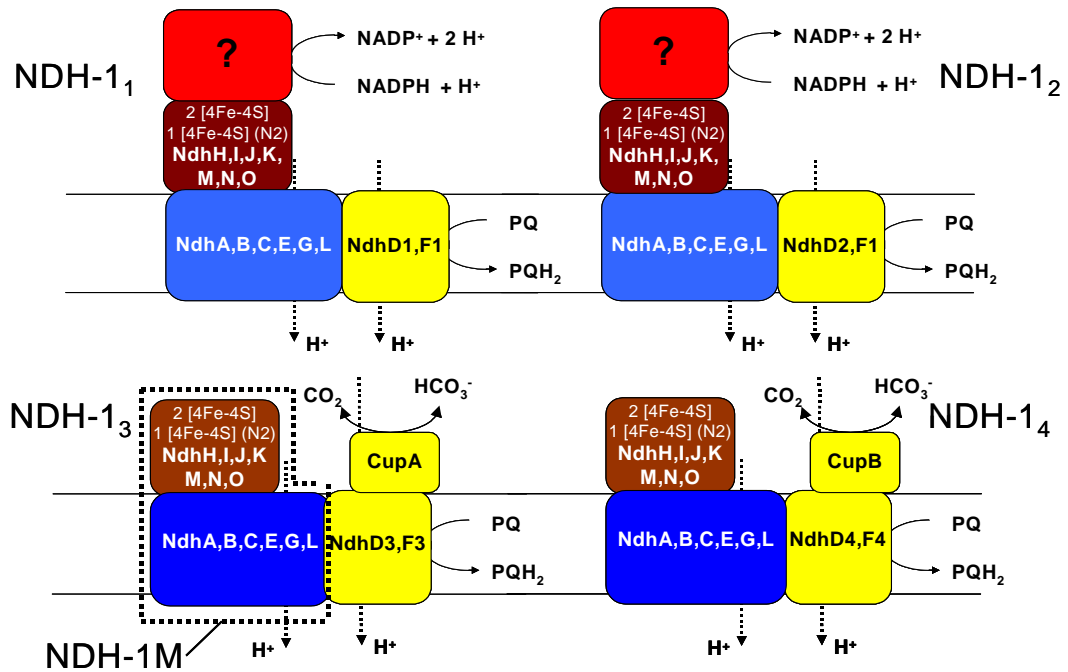


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Thanks: **T. Ogawa (M55), A Kaplan (Flv)**

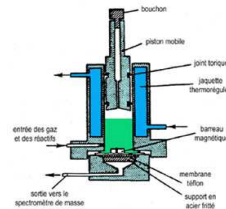
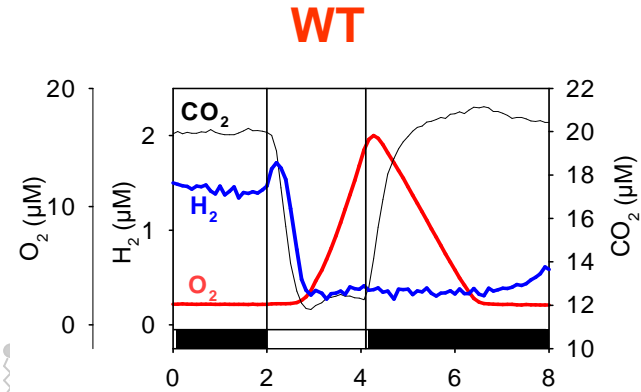
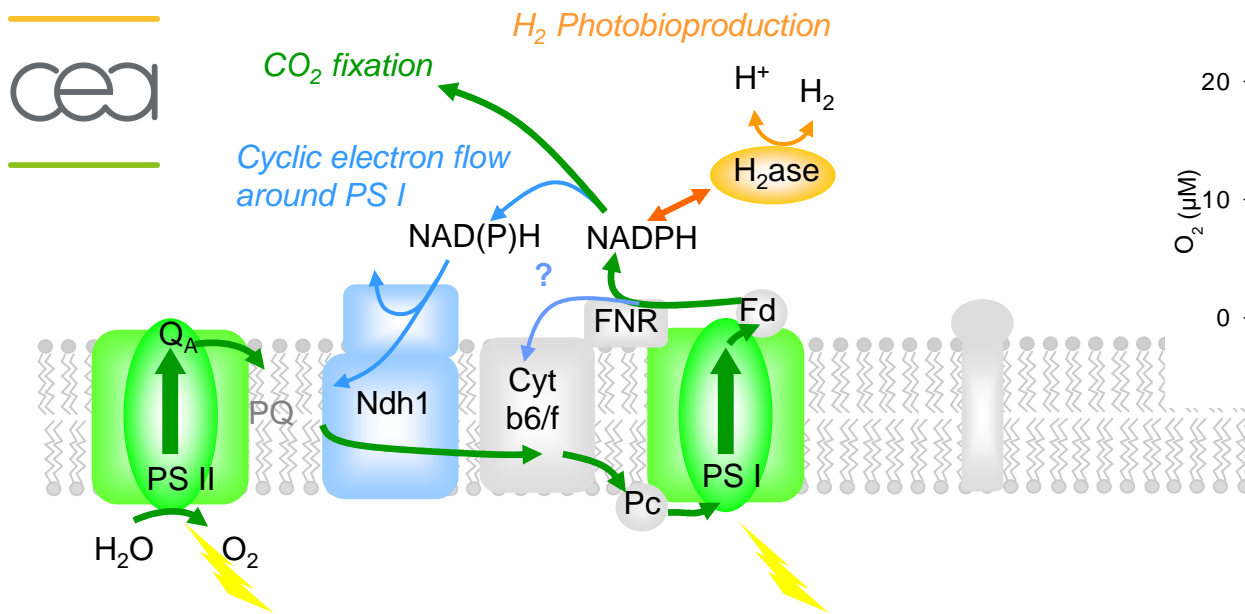


D1 and D2 (F1)
NAD(P)H oxidation

D3 and D4
CO₂ uptake

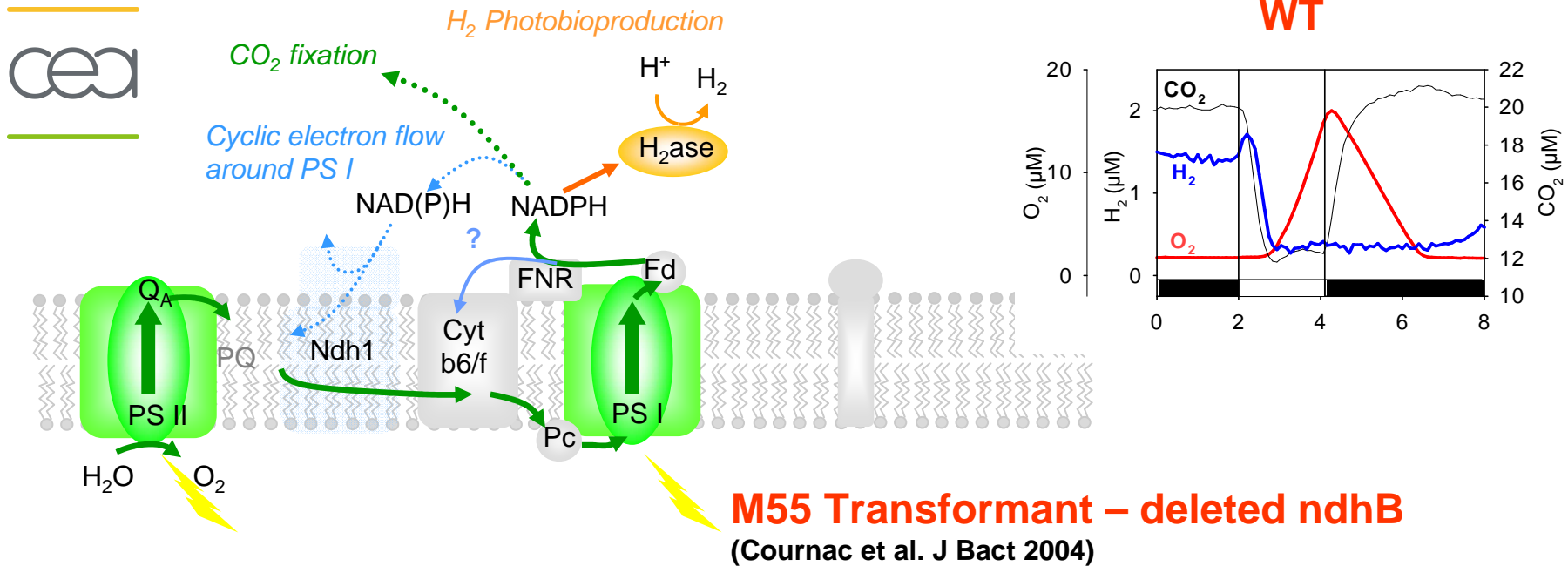
D5 and D6
??????????

H₂, CO₂ and O₂ exchange in *Synechocystis* PCC 6803



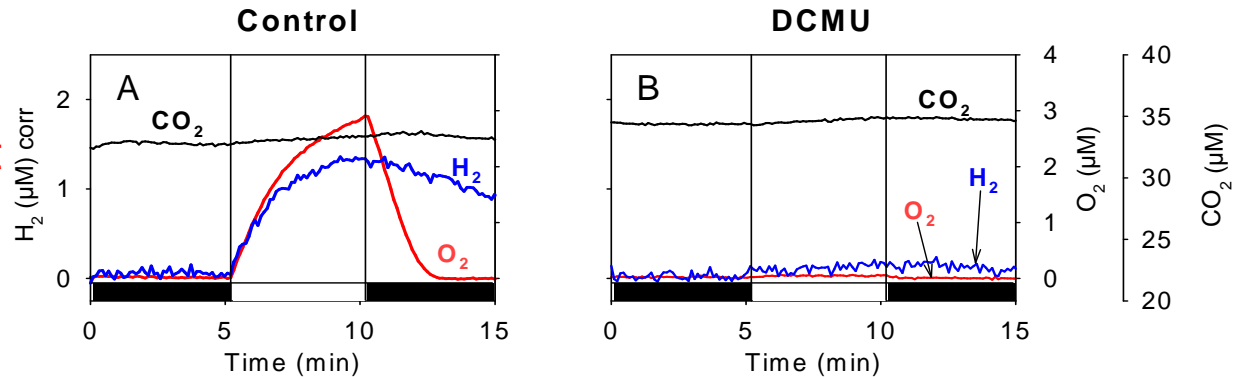
Liquid phase – Membrane inlet – Real-time online monitoring (H₂, O₂, CO₂)

H₂, CO₂ and O₂ exchange in *Synechocystis* PCC 6803

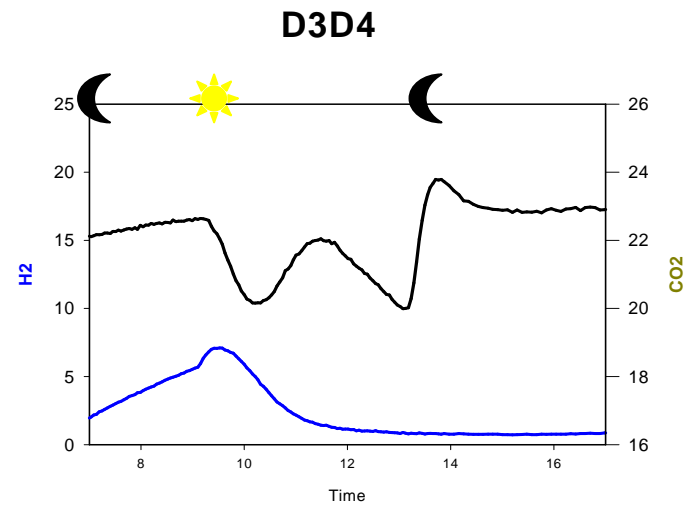
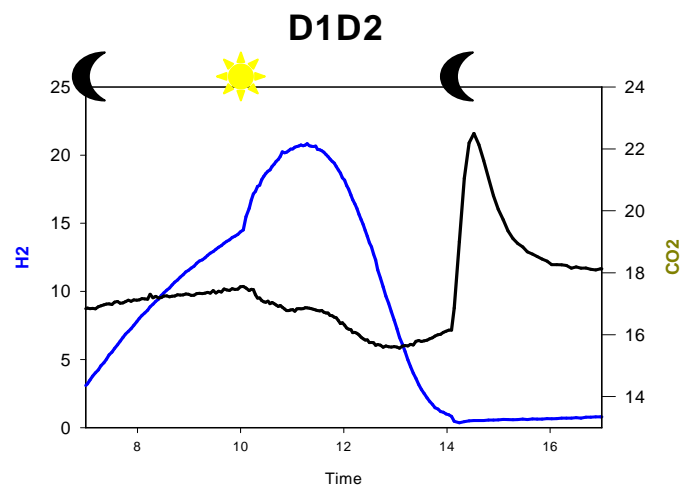
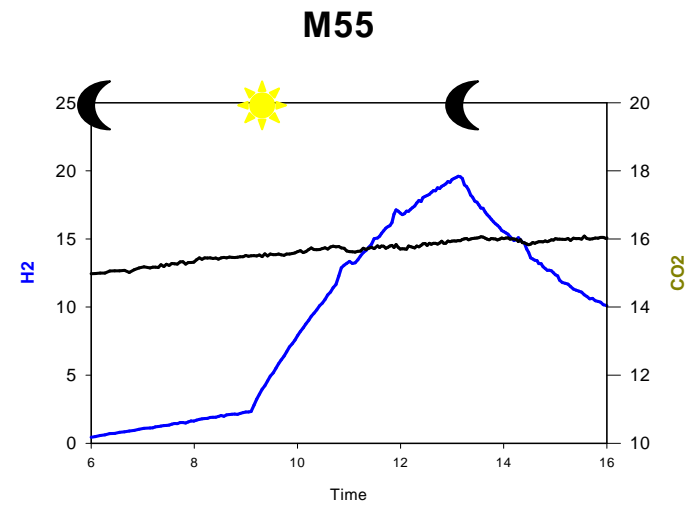
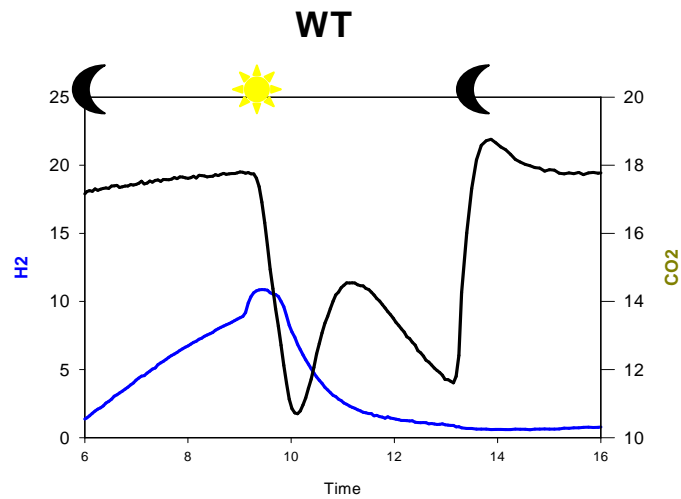


M55 Transformant – deleted *ndhB*
(Cournac et al. J Bact 2004)

Influence of the different NDH complex types?

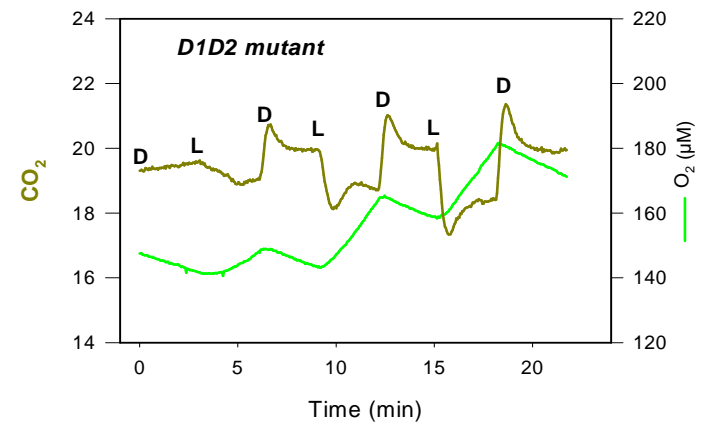
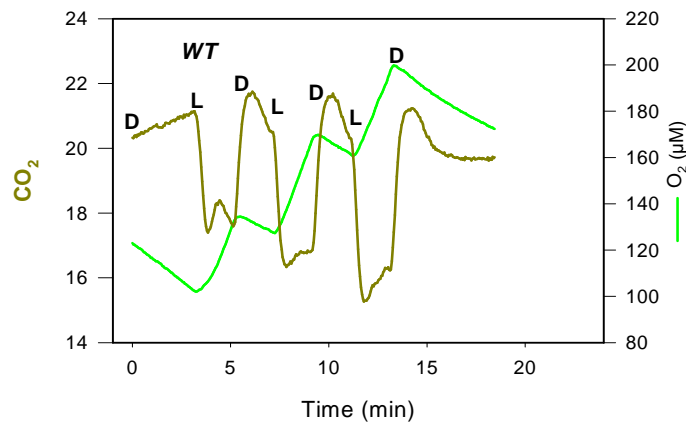


Hydrogen and CO₂ exchange in ndh mutants





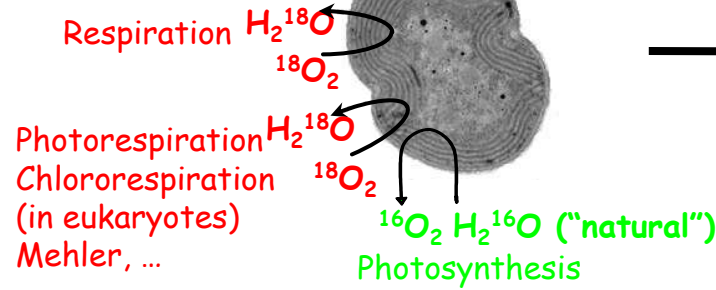
Delay in CO₂ uptake induction of dark-adapted $\Delta ndhD1,2$ mutant



Mass spectrometric analysis of photosynthetic and respiratory gas exchange – Isotopic tracing



Tracer = $^{18}\text{O}_2$

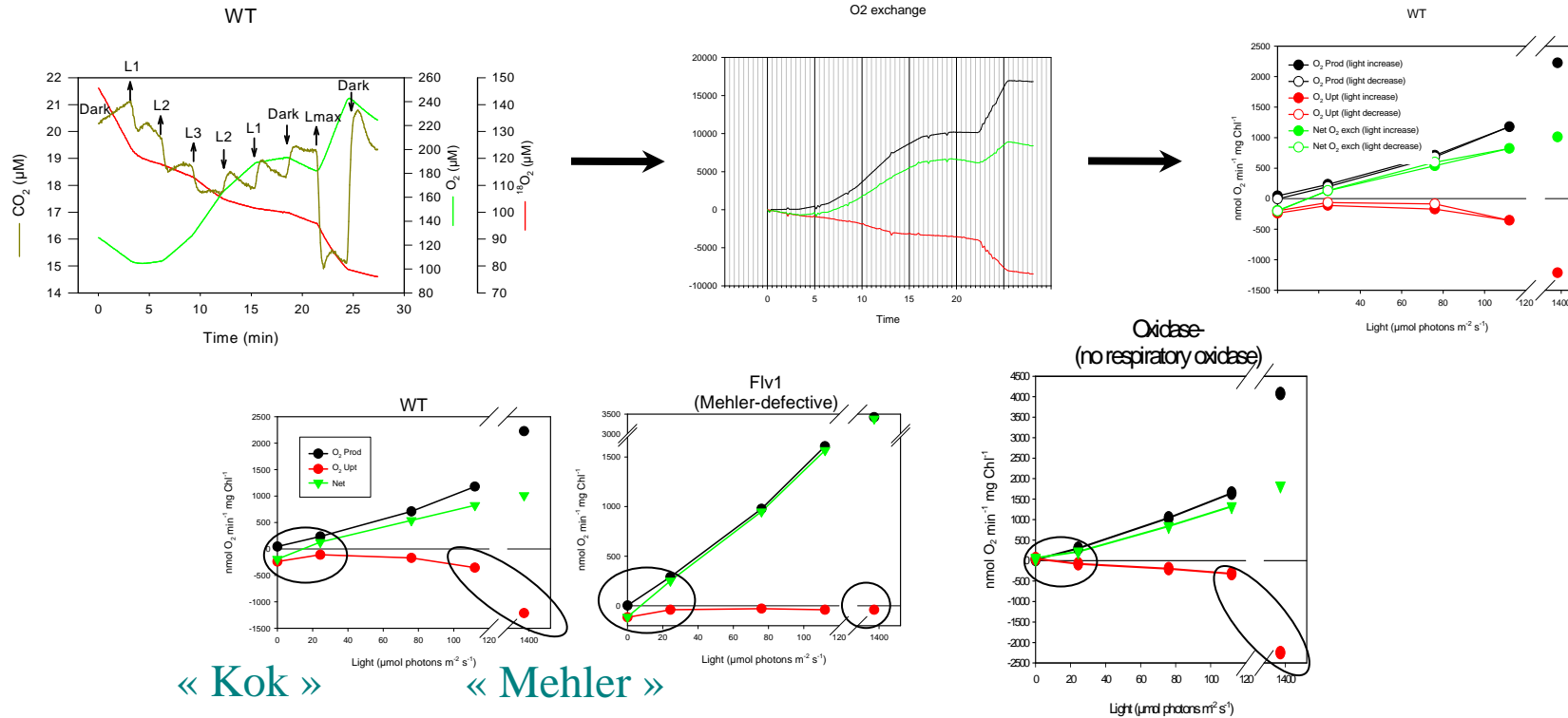


$^{16}\text{O}_2$: uptake, production
 $^{18}\text{O}_2$: uptake

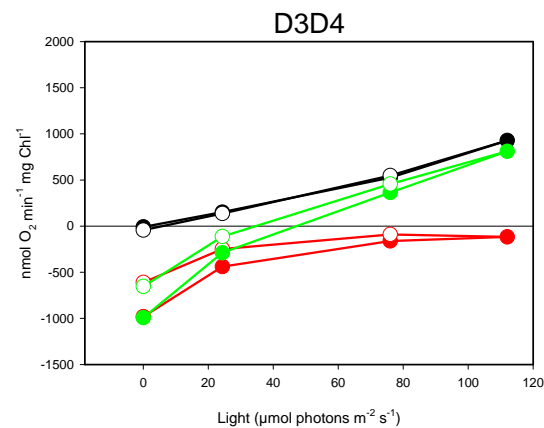
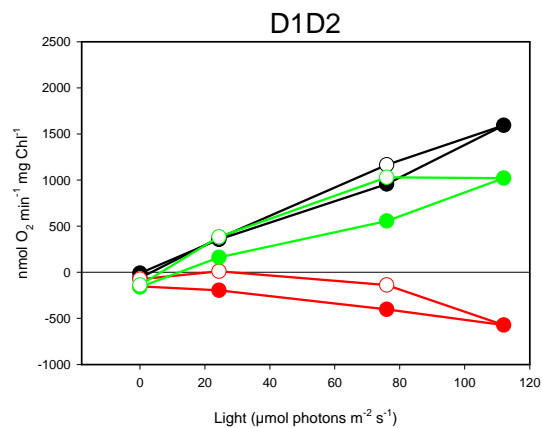
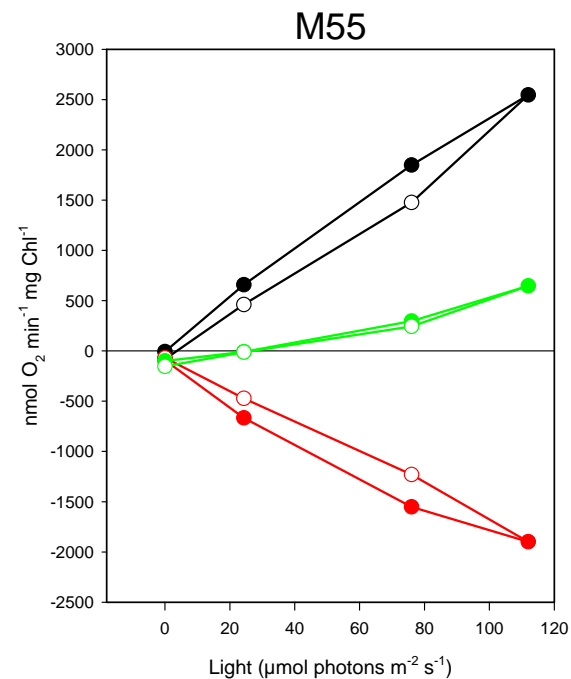
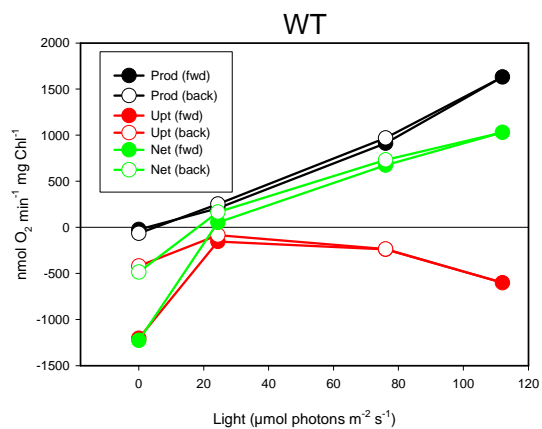
$$Uptake = -\frac{d^{18}\text{O}_2}{dt} \cdot \frac{^{16}\text{O}_2 + ^{18}\text{O}_2}{^{18}\text{O}_2}$$

$$Production = \frac{d^{16}\text{O}_2}{dt} + Uptake \cdot \frac{^{16}\text{O}_2}{^{16}\text{O}_2 + ^{18}\text{O}_2}$$

$$Net\ exchange = Production - Uptake = \frac{d^{16}\text{O}_2}{dt} + \frac{d^{18}\text{O}_2}{dt}$$



Light response of oxygen exchange rates in *ndhDx* mutants

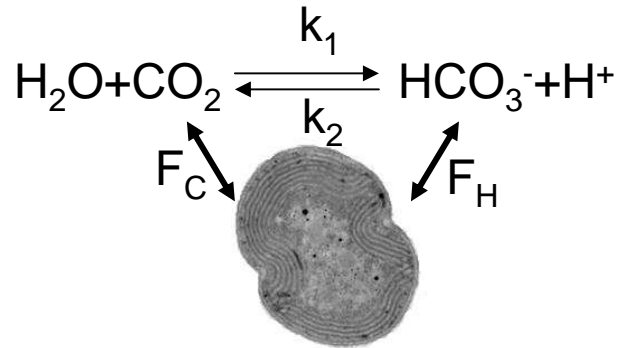


Mass spectrometric analysis of CO₂ and HCO₃⁻ exchange

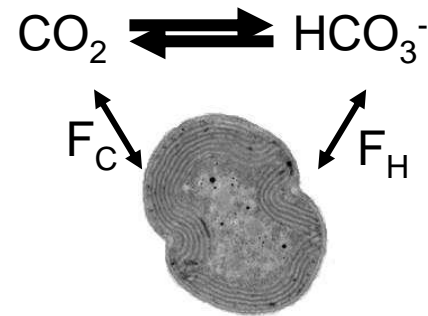
Carbonic anhydrase addition



No CA, slow equilibrium



+ CA, fast equilibrium



$$k_1 \approx 0.06 \text{ s}^{-1} \text{ at } 30^\circ\text{C}$$

$$k_2 = k_1 / (10^{\text{pH} - \text{pKa}})$$

$$[\text{HCO}_3^-] / [\text{CO}_2] = k_1 / k_2$$

WT

F_C and F_H equivalent with

$$C_t = [\text{HCO}_3^-]_{\text{-CA}} + [\text{CO}_2]_{\text{-CA}}$$

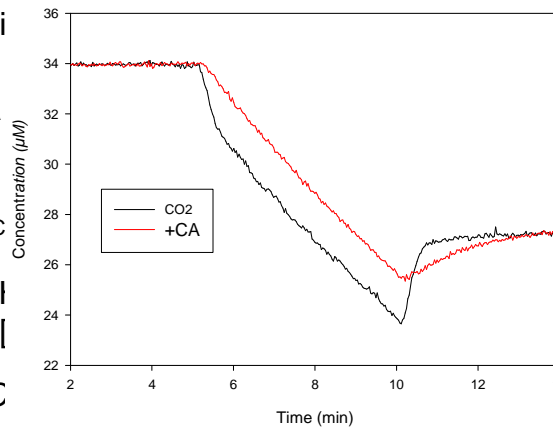
$$d(C_t)/dt = F_C + F_H$$

$$[\text{HCO}_3^-]_{\text{-CA}} = C_t - [\text{CO}_2]_{\text{-CA}}$$

$$F_C = d([\text{CO}_2]_{\text{-CA}})/dt - k_2 \cdot I$$

$$F_H = d([\text{HCO}_3^-]_{\text{-CA}})/dt + k_2 \cdot I$$

Possible difference in C



+10^{pH-pKa})

Impact fluctuations in F_C and F_H

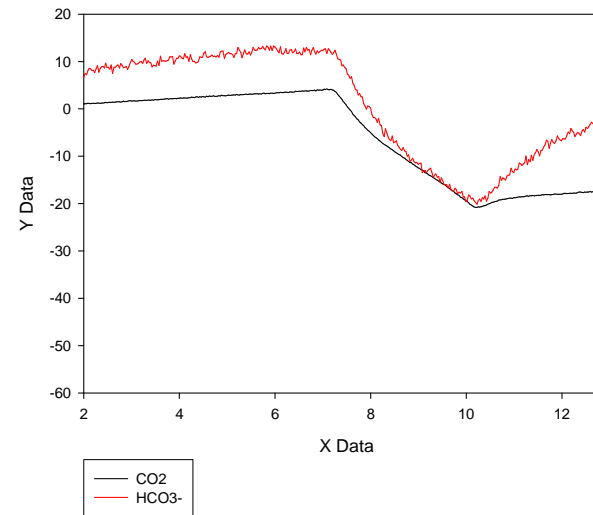
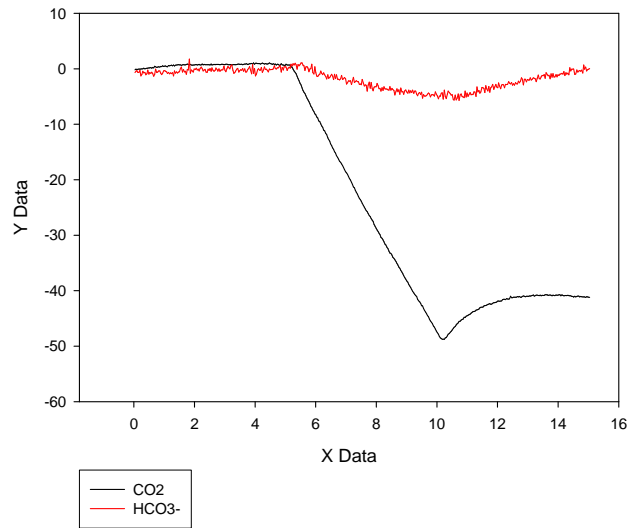
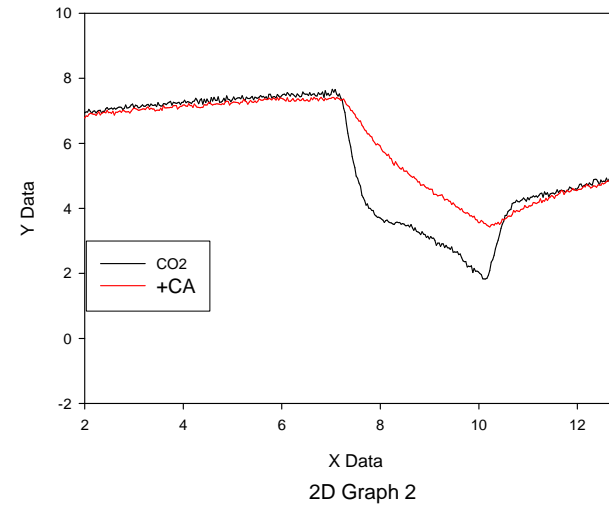
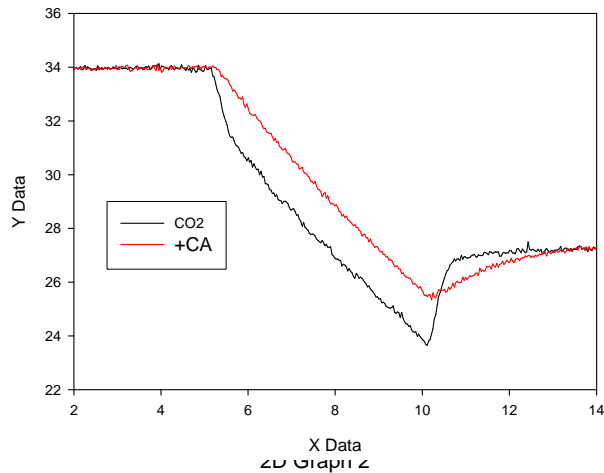
Observed concentrations – Reconstructed $\text{CO}_2/\text{HCO}_3^-$ fluxes



WT pH 7

(grown pH 8.3)

WT pH 7.5

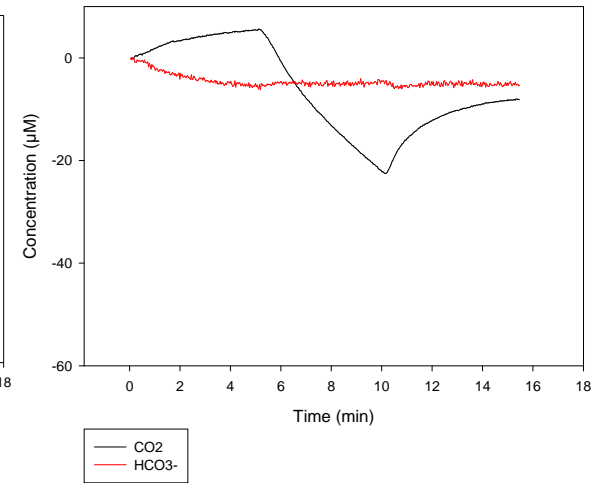
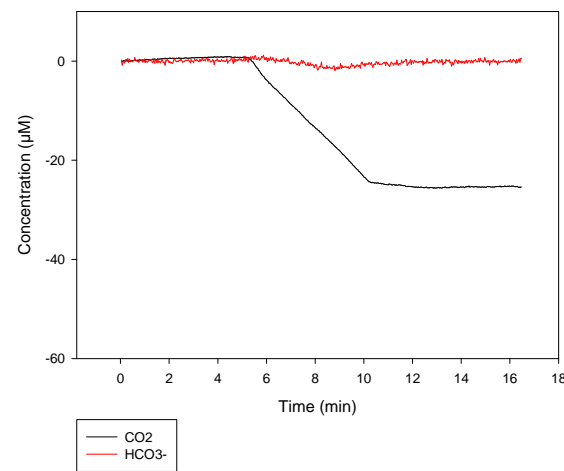
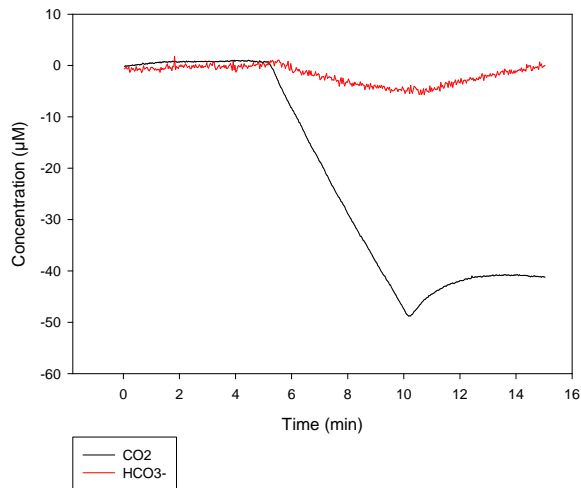
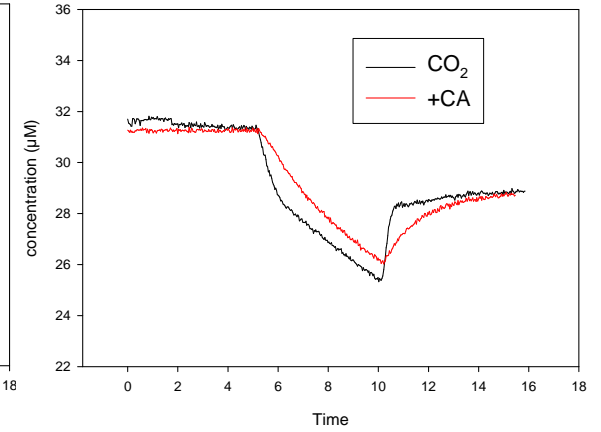
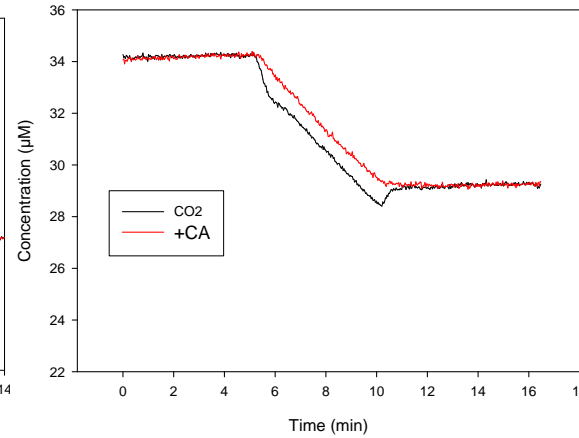
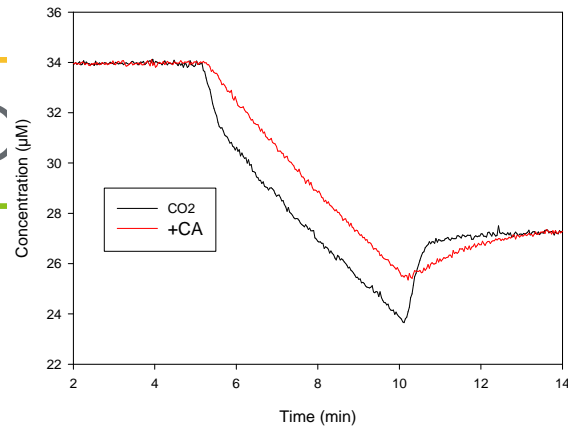




WT

D34

D12



CONCLUSIONS - QUESTIONS



- Deletions in *ndhD3/D4* slightly affect photosynthesis or H_2 exchange. A decrease in “Mehler” reaction rate has been observed. Link with Flv?
- Deletions in *ndhD1/D2* exhibit the stronger effects: as previously described, they affect respiration. In the light, O_2 uptake is stimulated (opposite to D3D4 mutant...): compensate for cyclic transfer impairment? Delayed induction of carbon uptake mechanisms and of photosynthetic activity (redox state-dependent?). In anoxia, H_2 production is stimulated.
- A new methodology for real-time estimation of CO_2 and HCO_3^- exchange rates has been set up and is to be challenged in future experiments.