Cellulose nanofibrils as a template for photosynthetic microbial biofuel production

M. Jämsä[a]*, S. Kosourov[a]*, V. Rissanen[b]*, M. Hakalahti[b], J. Pere[b], J.A. Ketoja[b], T. Tammelin[b]**, Y. Allahverdiyeva[a]**

[a] Molecular Plant Biology, Department of Biochemistry, University of Turku, Turku, FI-20014 Finland
[b] VTT Technical Research Centre of Finland, VTT, PO Box 1000, FIN-02044 Espoo, Finland
* These authors contributed equally
** corresponding authors:
Assoc. Prof. Yagut Allahverdiyeva, e-mail: allahve@utu.fi, Molecular Plant Biology, Department of Biochemistry, University of Turku, FI-20014, Turku, Finland
Dr. Tekla Tammelin, e-mail: tekla.tammelin@vtt.fi, VTT Technical Research Centre of Finland LTD, VTT, PO Box 1000, FIN-02044 Espoo, Finland

Electronic supplementary information (ESI)

Quartz Crystal Microbalance with Dissipation Monitoring (QCM-D)

In QCM-D a piezoelectric quartz crystal sensor oscillates under a pulsating electric field at a specific fundamental resonance frequency f_0, and its overtones,[1] which depends on the total oscillating mass. If the layer covering the sensor is even in distribution, rigidly attached, fully elastic and has a small mass compared to that of the sensor crystal, a change in resonance frequency is directly proportional to the change in areal mass according to the Sauerbrey equation (eq 1):[2]

$$\Delta m = -C\frac{\Delta f}{n} \quad (1)$$

where Δm is the mass change per unit surface, $\Delta f = f - f_0$ is the change in resonance frequency, C is the sensitivity constant of the sensor and n is the measurement overtone number (n=1, 3, 5, 7, 9, 11). Simultaneously, when the voltage is periodically cut off, the oscillation gradually decreases and the resonance amplitude dampens due to frictional losses in the film layer. This dissipation of energy, D, represents the viscoelastic properties of the sample, and can be presented as:

$$D = \frac{E_{disspation}}{2\pi E_{storage}} \quad (2)$$

where $E_{dissipation}$ is the dissipated energy and $E_{storage}$ is the total energy stored during one oscillation cycle. The change in dissipation, ΔD, is a qualitative measure of the rigidity and softness of the layer adhered to the sensor surface. The film can be considered fully elastic and rigid when $\Delta D \leq 1 \times 10^{-6}$, and the overtones of Δf and ΔD do not spread significantly.

Preparation of supported TEMPO CNF thin films

TEMPO CNF suspension was diluted to 0.15 wt% concentration using Milli-Q water and sonicated with a Branson Digital Sonifier (400 W, 20 kHz) using a 25 % amplitude for 2 min. Before deposition the QCM-D sensor surfaces were cleaned using UV/ozone (Bioforce Nanosciences, CA), rinsed with Milli-Q water and dried with N_2 gas. The sensors were then coated with 200 µl of 1 mg ml-1 PEI solution in Milli-Q water by drop casting for 30 min to ensure the anchoring of the fibrils, rinsing with water and drying with N_2 gas. Then 200 µl of TEMPO CNF solution was dispensed onto the sensors and spin-coated (WS-400BZ-6NPP/Lite, Laurell, North Wales PA, USA) at 3000 rpm for 2 min, followed by heat treatment for 10 min in 80°C oven to ensure fibril attachment. The surfaces were stored in desiccator in darkness prior to QCM-D measurements.

Areal mass determination of attached Δ*hupL* filaments on TEMPO CNF thin films

Prior to the measurements, all surfaces were stabilized in desiccator for at least 30 minutes in order to ensure a constant moisture content of the thin films. A baseline was determined by measuring the frequency response of plain TEMPO CNF coated sensor crystal surface for 30 minutes. The frequency data was similarly collected again after cyanobacterial filaments attachment experiment using exactly the same sensor. Finally, the data was stitched

together with the QTools software and the areal mass was calculated from Δf according to the Sayerbrey equation. The data collected using the seventh overtone (35 MHz, f0 = 5 MHz, n = 7) was used in the areal mass calculations.

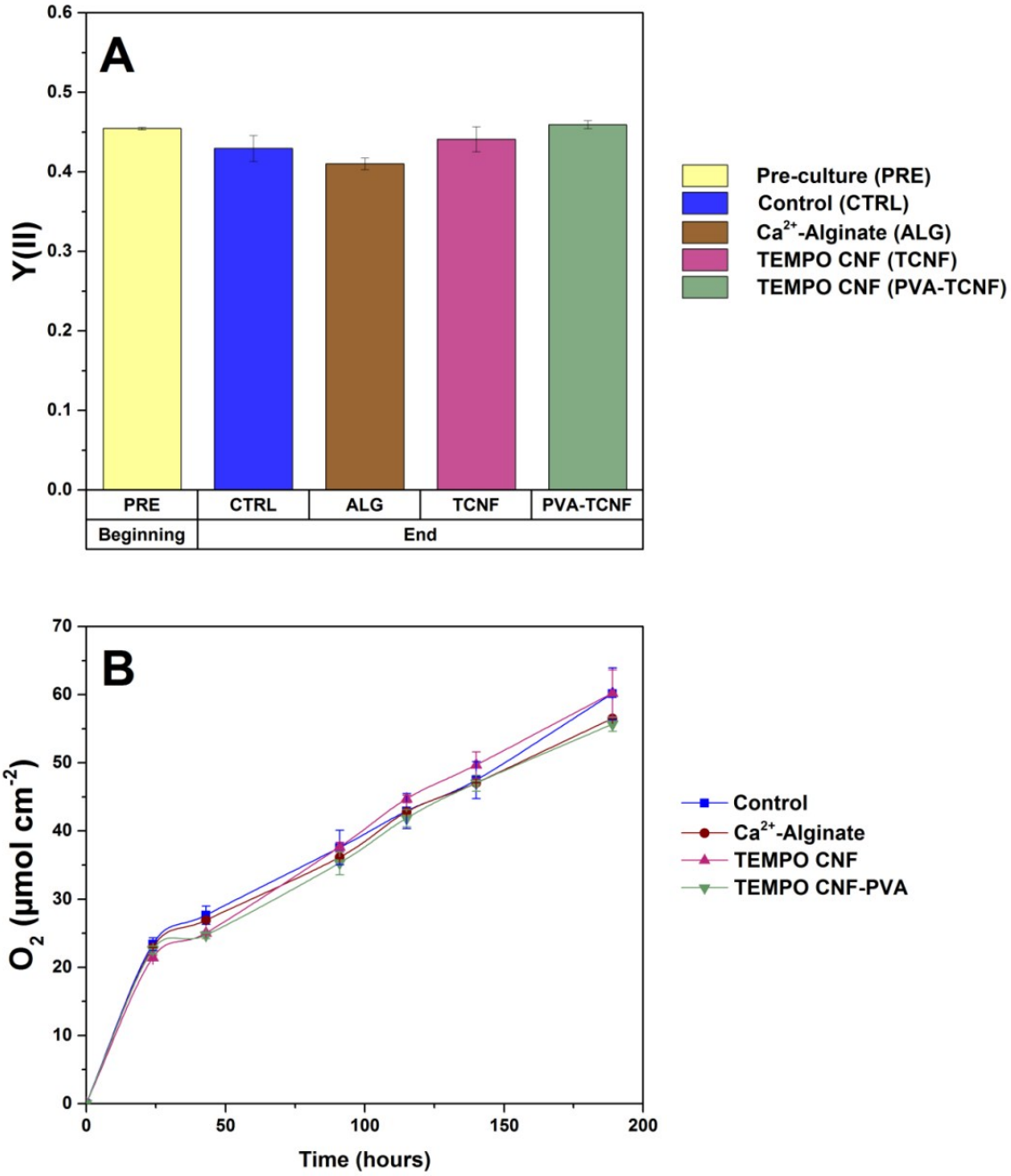

Figure S1. The photosynthetic activity (A) Y(II) of the cyanobacterial cells in pre-culture and after 189-hour hydrogen production stage when approach A has been used. Oxygen evolution (B) during the hydrogen production stage.

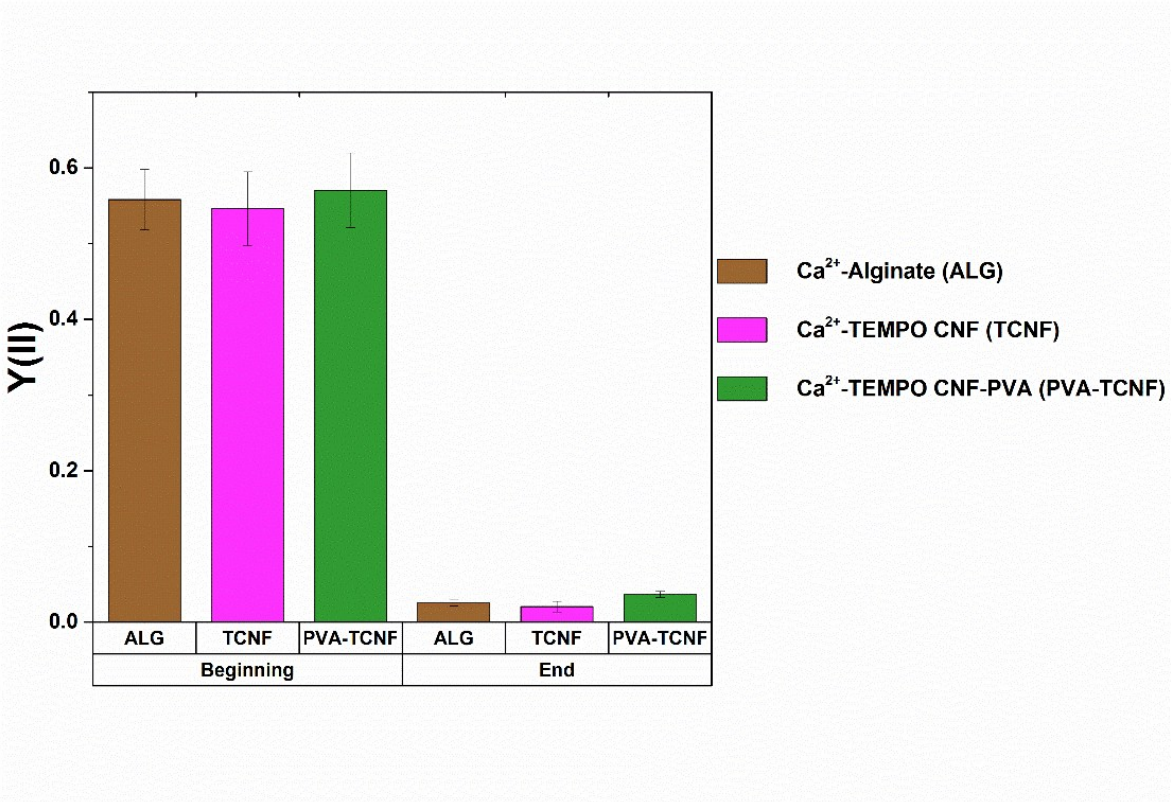

Figure S2. The photosynthetic activity Y(II) of the sulfur-deprived *C. reinhardtii* cells entrapped in different Ca^{2+}-hydrogel layers in the beginning (0 h) and in the end (312 h) of the H_2 photoproduction experiment.

Table S1. Chlorophyll a content of cyanobacterial strips used in approaches A and C measured as described in materials and methods complementary analytical methods section.

		Beginning		
		Chl a (µg/cm²)		
Approach A	Control	33 ± 16		
	Ca^{2+}-Alginate	29 ± 2.7		
	TEMPO CNF	28 ± 1.2		
	TEMPO CNF-PVA	24 ± 2.1		
		Beginning	After recovery	
		Chl a (µg/cm²)	Chl a (µg/cm²)	Recovery-%
Approach C	Control	14 ± 3.3	7.7 ± 0.53	-45 ± 3.8
	Ca^{2+}-Alginate	10 ± 1.7	11 ± 1.4	10 ± 14
	TEMPO CNF-PVA-1	11 ± 0.89	8.4 ± 2.0	-24 ± 18
	TEMPO CNF-PVA-2	12 ± 1.4	14 ± 1.0	16 ± 8.3

References

1 F. Höök, M. Rodahl, P. Brzezinski and B. Kasemo, *Langmuir*, 1998, **14**, 729–734.

2 G. Sauerbrey, *Zeitschrift für Phys.*, 1959, **155**, 206–222.