# Mechanical Characterization of a Single *Synechocystis* sp. Strain PCC 6803 Cell in Different Osmolarity Solutions


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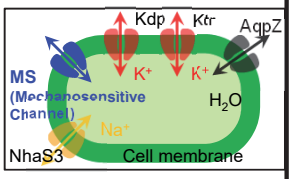
## Stiffness Measurement of a 2 μm Cell Using On-chip Force Sensor

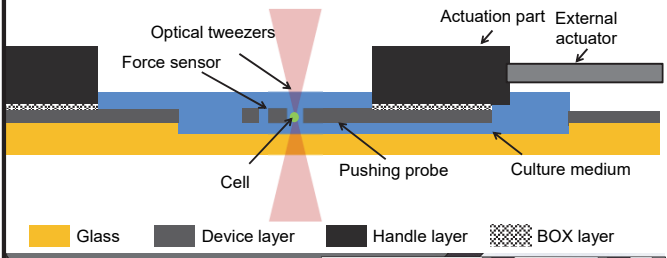
## Background & Purpose

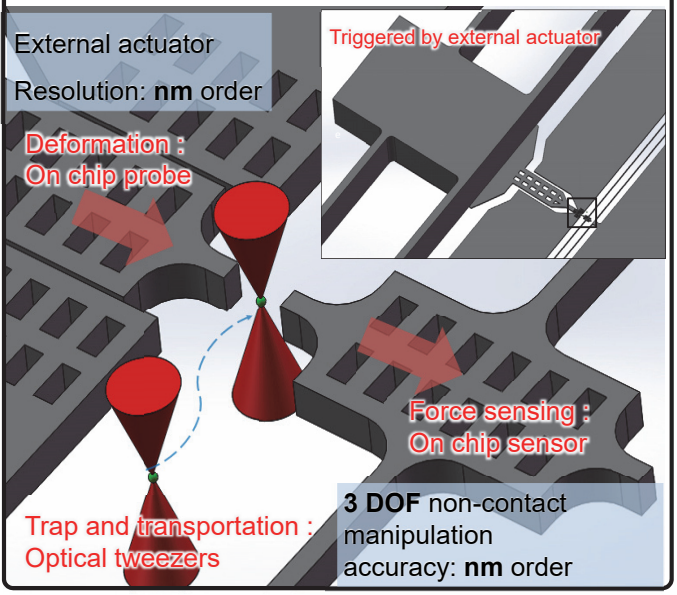
***Synechocystis* sp. PCC6803**
**Floating unicellular** cyanobacterium
Size : **2 μm**
**Model** microorganism for the study of **photosynthesis**, **biofuel** and **acclimation** to environmental changes.



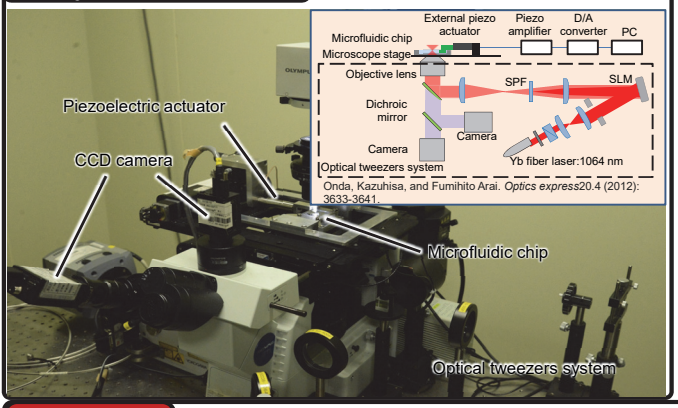

**Purpose** : Evaluate the mechanical properties of wild type (WT) and mutant type lack of mechanosensitive channel (*ΔmscL*) cell in different osmolarities

## Concept of Microfluidic chip



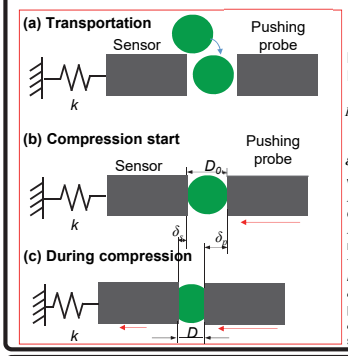




## System Setup




## Measurement method

### Cell Compression




$F_s = k\delta_s$

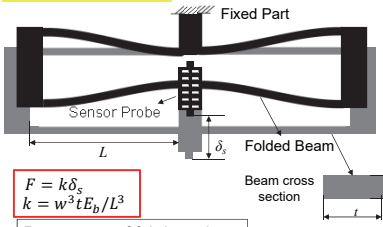
By applying Hertzian model

$F_s = \frac{E_c \varepsilon^{3/2}}{3(1-\nu^2)} D_0^2$

$\varepsilon = (\delta_p - \delta_s)/D_0$

where
$D_0$ is cell's original diameter.
$E_c$ is cell's Young's modulus.
$\nu$ is Poisson's ratio
$k$ is spring constant
$\delta_p$ : displacement of pushing probe
$\delta_s$ : displacement of sensor

### Force sensor




$F = k\delta_s$
$k = w^3 t E_b / L^3$

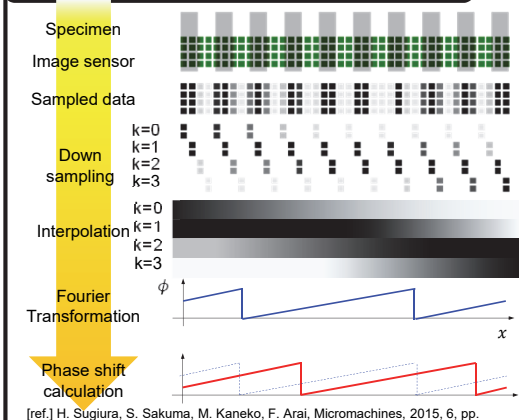
Parameters of fabricated force sensor:

| | | |
|---|---|---|
| $w$ | = | 4.5 μm |
| $t$ | = | 7.5 μm |
| $L$ | = | 1500 μm |
| $E_b$ | = | 170 GPa |

Thus, theoretical spring constant :

**$k$ = 0.034 N/m**

## Sampling Moiré Method




[ref.] H. Sugiura, S. Sakuma, M. Kaneko, F. Arai, Micromachines, 2015, 6, pp. 660-673, 2015

$I_k(x) = I_a \cos\left(\phi_m + k\frac{2\pi}{N}\right) + I_b$

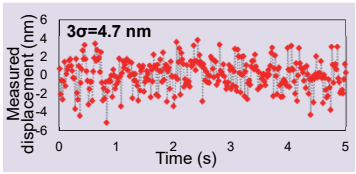
DFT

where
$I_a$: : amplitude of grating intensity
$I_b$ : background intensity
$\phi_m$ : phase of moiré fringe

$\phi_m(x) = -\tan^{-1}\left(\frac{\sum Isin\left(k\frac{2\pi}{N}\right)}{\sum Icos\left(k\frac{2\pi}{N}\right)}\right)$

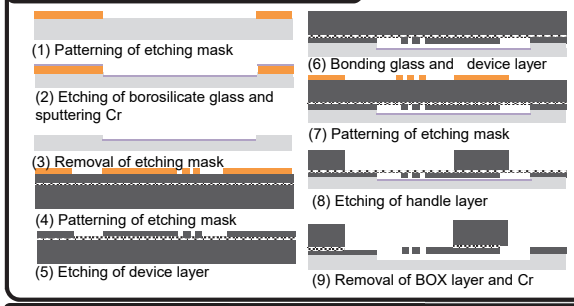
displacement :
$u = \frac{\phi_m - \phi_m(t=0)}{2\pi} p$

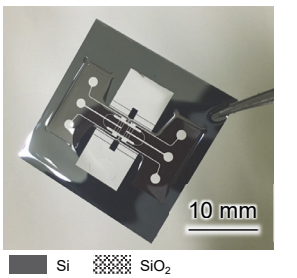
where
$p$ ; the pitch of the gratings




Measurement of reliability when the probe was fixed.

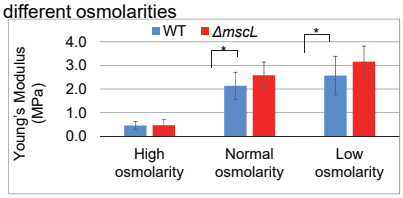
## Fabrication process



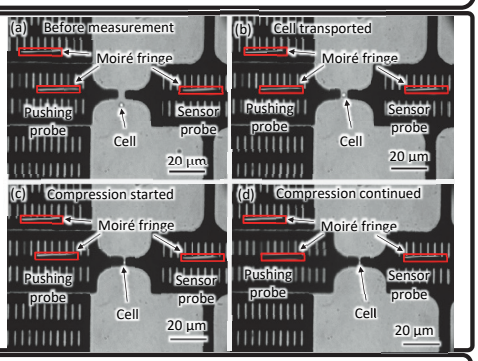

## Result

Measurement result of WT and *ΔmscL* in three different osmolarities




Error bars stand for standard deviation and a significant difference between WT and *ΔmscL* was analyzed by Student's t test (* $p < 0.05$).

High osmolarity: BG11+1mol/L sorbitol
Normal osmolarity: BG11
Low osmolarity: BG11:DI water = 1:9




## Reference


1. Chang, D., Sakuma, S., Kera, K., Uozumi, N., & Arai, F. (2018). Measurement of the mechanical properties of single Synechocystis sp. strain PCC6803 cells in different osmotic concentrations using a robot-integrated microfluidic chip. Lab on a Chip, 18(8), 1241-1249.
2. Chang, D., Sakuma, S., Kera, K., Uozumi, N., & Arai, F. (2017, December). Mechanical characterization of a single synechocystis sp. PCC 6803 cell in different osmolarity solutions. In Micro-NanoMechatronics and Human Science (MHS), 2017 International Symposium on (pp. 1-3). IEEE.



**Abstract:** *Synechocystis* sp. strain PCC6803 (*Synechocystis*) is a model microorganism and its mechanosensitive (MS) channels play important roles in its osmoadaptation mechanism. When the osmotic concentration of the culture environment changes, the inner pressure of the cell also changes due to the transportation of water through ion channels. Because the tension in the cell membrane relates to the inner pressure, we expect that the response of the MS channels to an osmotic concentration change could be evaluated by measuring their mechanical properties. Here, we propose a system for the measurement of the mechanical properties of a single *Synechocystis* cell. We developed a robot integrated microfluidic chip combined with optical tweezers. The chip has an external actuated pushing probe and a force sensor probe. A single cell was located between the tip of both probes using the optical tweezers and was then deformed using the probes. As a result, we could measure the force and deformation and compare the Young's moduli of two groups; a group of wild type cells and a group of mutant (genetically modified) cells with a defect in the MS channels, at three different osmotic concentrations. The results showed that the Young's modulus of each group changed according to the osmotic concentration, while changes in cell size were too small to be detected. These results confirmed that the proposed evaluation method provides an understanding of the physiological function of MS channels for keeping the cell integrity of microorganisms when the cells are exposed to different external osmotic changes.