

Book of Abstracts related to the number of filtered particles. The expelling phase was occurred in between the ingestion phase and released the filtered particles toward the ex-current channels. Consequently, the particles expelled through the osculum for a clearance in the choanochambers.

The multiple sponges made a complex flow structure surrounding their bodies even in still condition as shown in Fig.2. These in-current and ex-current flows made chaotic circulating flow around them. It would enhance the mixing of surrounding fluid, especially lateral mass transportation. The gregariousness of sponges might produce some benefits on the transportations of nutrition, bacteria, and oxygen by the extrusion from the osculum and the intake from the whole surface of sponges.

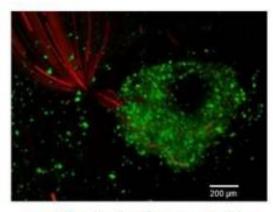


Fig. 1. Visualization of ex-current and incurrent flows and filtered particles of sponge

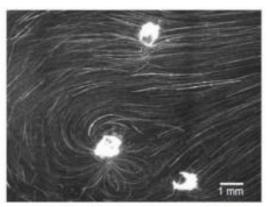


Fig. 2 Visualization of ex-current flows generated by multiple sponges in still dish.

[Fig. 1-2]

# 11.1.2 Hydrogel nanofilaments in oscillatory microchannel flow

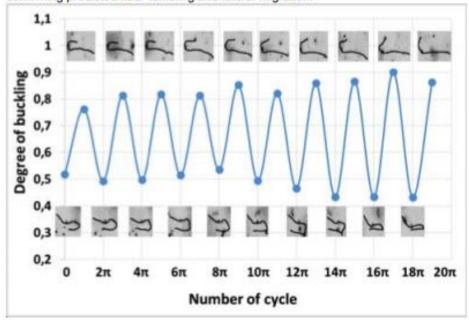
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geometry for conducting targeted material delivery to regenerate aligned tissues or perform DNA transport. Hence, in the following we analyze transport properties of such objects conveyed by oscillating flow simulating typical extracellular environment. Due to nanometric size of our nanofilaments both hydrodynamic interactions and Brownian fluctuations have to be considered. The problem has analogy to exhaustive studies of DNA flow dynamics, though the coarse filaments material allows to limit problem description to purely hydrodynamic interactions, neglecting complex molecular and electrostatic interactions. We report for the first time ever the use of hydrogel nanofilaments as an alternative to model flow dynamics of molecular chains. FITC-fluorescently labeled hydrogel nanofilaments were prepared by co-axial electrospinning of two immiscible polymers (PLCL polymer and NIPAAm/BIS-AAm hydrogel solution), with the former one being used to mold the hydrogel in the core-shell structure [1]. After removal of their shells, hydrogel nanofilaments suspended in water - DMF (4:1) solution were introduced into the PDMS microchannel using a syringe micro-pump. The microchannel was equipped with two additional pre-chambers used to collect nanofilaments and to introduce the selected one into the observation area. Care was taken to analyze nanofilaments solely remaining in the plane of observations. In-house designed squeezetube micro-pump was used to produce sinusoidal oscillating flow within the channel. The velocity amplitude V<sub>max</sub> varied from 100 µm/s to 900 µm/s, flow oscillations frequency was set around 0.1 Hz. Nanofilaments were imaged using epifluorescence microscope (Leica AM TIRF MC) equipped with 20x/0.40 NA air objective and a mercury lamp light source. The flow-induced migration of nanofilaments was recorded using a high-gain EM-CCD camera (C9100-2, Hamamatsu) with typical frame rate of 15 Hz. The mechanical properties of our nanofilaments such as persistence length (Lp) and bending stiffness were evaluated from their Brownian shape deformations. Selected for the experiment nanofilaments were characterized by flexural modulus of 2 kPa, typical radius R of about

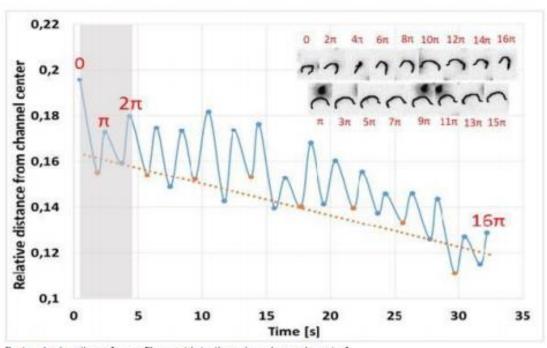
Recently, we demonstrated possibility to produce hydrogel in a form of nanofilaments [1], the supreme

50 nm, and contour length L of several micrometers.

Performed experimental investigations demonstrated presence of lateral migration of nanofilaments, and their complex bending dynamics, being characteristic for long biomolecules. Experimental data are compared with hydrodynamic worm-like beads model of fibers conveyed by shear flow [2], confirming predicted fiber tumbling and lateral migration.



[The buckling parameter of the filament evaluated at each phase of oscillatory flow.]



[Lateral migration of nanofilament into the microchannel center]

Our electrospun nanofilaments offer a unique capability to mimic dynamics of long flexible object conveyed by a hydrodynamic flow. At the same time, they can be successfully used to validate essential assumptions of computational models used to describe dynamics of complex suspensions [3].

### Acknowledgments: NCN grant 2015/17/N/ST8/02012

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- Lateral migration of electrospun hydrogel nanofilaments in an oscillatory flow, S. Pawlowska, P. Nakielski, F. Pierini, I.K. Piechocka, K. Zembrzycki, T.A. Kowalewski, PLOS ONE 12(11):e0187815 (2017)
- 3. Flow of complex suspensions, A. Lindner, Phys. Fluids 26:101307 (2014).

#### 11.1.3

## Visualizing the swim technique of a puller-type micro-swimmer without visualizing the flagellum

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Introduction: The locomotion mechanism of *Euglena gracilis* is investigated using microscopic shadow imaging and Micro Particle Image Velocimetry (µPIV). Three distinct locomotion modes were observed; translation, spin and left/right turn. Since the flagellum was not possible to image, the strokes were identified by evaluating the flow field around the protist. The flow field information is obtained using a phase-separated PIV evaluation, which uses of a histogram-thresholding based dynamic masking approach. The temporal resolution of the experiment was sufficient to identify the sequence of translation and spin, and the stroke-pulling frequency. The results indicate that the organism has a complex locomotion technique that allows the change of direction, change of axial orientation and propulsion.

Euglena gracilis (E. gracilis) is a protist microorganism that populates in fresh water habitats and sustains life by collecting solar energy by photosynthesis using chloroplasts. Its body length without the flagellum can vary between 20 μm and 100 μm. The optimum light level for chloroplasts is around 30 W/m², and therefore E. gracilis is known to adjust its depth in water in order to find this optimum light condition. This necessitates the photo detection, sense of gravity and locomotion, which are vital functionalities for E. gracilis' survival. In the full paper, we will provide a brief summary of E. gracilis locomotion behaviour, which consists of thee modes; forward translation, spin along long axis and the euglenoid movement.

**Methods:** The experiments were performed in still water and a cold light source ensured that there was no background flow due to convection currents. The flow field around *E. gracilis* is measured using time-resolved MicroPIV. The MicroPIV system components include an inverted fluorescence Microscope, a sensitive CMOS detector, a synchronization device, and a pulsed LED illumination system. In biological flows, high-power pulsed laser illumination is not preferred as this can disable the organism. For this reason, a lower-power, green LED-based pulsed illumination was used in transmission mode.

**Dynamic masking:** Before PIV evaluation, the image of *E. gracilis* was removed from the flow field using dynamic masking. This is necessary because the movements of the organism are not related to the fluid motion in the vicinity of *E. gracilis*. If not removed, the moving features on *E. gracilis* image (stigma, nucleus etc.) contributes to the cross correlation function during the velocity calculation and introduces an error in of the flow field. The details of the image pre-processing steps used to achieve the dynamic mask are provided in Ergin (2017).

Velocimetry: Details of the velocity calculations are provided in Ergin (2015). Briefly, an adaptive PIV algorithm is used, which is an advanced particle displacement estimator implemented in DynamicStudio (Dantec Dynamics, Skovlunde, Denmark). The calculation is a cross-correlation based, adaptive and iterative procedure with vector validation and deforming windows. For the current case, interrogation windows of 32x32 pixel are used with 75% overlap. Window deformation is performed by adapting the interrogation area shape to velocity gradients. Several passes can be made to further shift & deform the windows to minimize the in-plane particle dropout. This procedure is repeated until a convergence limit in pixels or a maximum number of iterations is reached. Then a 2D Gaussian fit is performed on the highest correlation peak to calculate the displacement field with sub pixel accuracy. Between passes, spurious vectors are identified and replaced with a number of validation schemes. The sub pixel positioning accuracy of the Adaptive PIV algorithm is reported as 0.06 pixels with 95% confidence (Ergin et al. 2015). The 0.06 pixels correspond to a 27.5nm