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Biophysics of green algae: *Euglena gracilis* investigated by atomic force microscopy

C. Gruenberger[1,a], R. Ritter[1], F. Aumayr[1], H. Stachelberger[2], I. C. Gebeshuber[1,3]

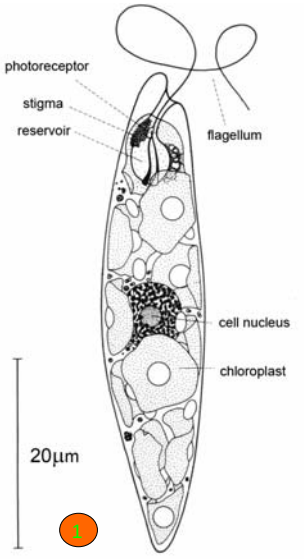
[1] Institut für Allgemeine Physik, TU Wien, Wiedner Hauptstrasse 8-10/134, 1040 Wien, Austria
[2] Institut für Verfahrenstechnik, Umwelttechnik und Technische Biowissenschaften, TU Wien, Getreidemarkt 9/166, 1060 Wien, Austria
[3] AC²T Austrian Center of Competence for Tribology, Viktor Kaplan-Strasse 2, 2700 Wiener Neustadt, Austria
Please send correspondance to [a]gruenberger@iap.tuwien.ac.at

Overview

Materials produced by living organisms are remarkable in many ways. Their **functional structures** simultaneously solve various optimization problems at different length scales, leading to nanometer-precision and highly integrated, miniaturized and robust components. Applying knowledge about these principles to **technical innovations** requires sound understanding also at the nanometer scale. Our model organism, the green alga species ***Euglena gracilis***, comprises a range of interesting bionanomaterials, for example flexible yet tough cell walls, compact crystalline energy storage sites and a thermally stable high-yield single photon detector [1,2,3]

Material and methods

Atomic force microscopy (AFM) allows for investigation of conductive and non-conductive samples in various environments (such as in ambient conditions or in liquid) with unprecedented resolution.
The possibility of acquiring additional information, e.g. on viscoelastic properties, and performing micromanipulations on the biomaterial made AFM the scanning technique of choice. Our MFP-3D atomic force microscope (Asylum Research, Santa Barbara, CA, USA) is equipped with top-view optics and an inverted microscope enabling investigation of transparent as well as opaque samples. Preparation techniques for **whole cells and cell parts** were developed, imaging was performed in alternating contact mode in air at a scan rate of 0.1 Hz.



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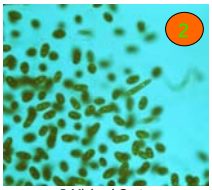
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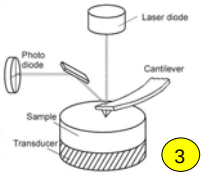

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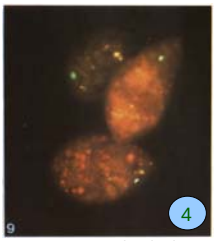
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The green alga *Euglena gracilis*

Euglena gracilis is a single-celled freshwater organism with a **typical length of 20µm – 100µm**. The single cells move through water by a rod-like structure, the flagellum. Orientation of the cells is controlled by a simple but complete visual system, consisting of the **photoreceptor**, the stigma and the **flagellum**. During its movement, the cell rotates and the light intensity on the photoreceptor varies periodically, causing an arbitrary change in the direction of movement, until this variation no longer occurs and the cell is moving towards the light source.
The **pellicle**, the outer shell, gives mechanical stability to the cell being highly flexible nonetheless. Interlocking pellicle strips slide against each other during cell movement. This movement is lubricated by **biogenic lubricants** (mucus) excreted by pellicle pores.

Results

A **preparation technique for whole dried intact cells** was developed. **AFM** images of **whole cells** show the **pellicle**, **mucus excretion pores** and **new surface features**, not yet reported in the scientific literature.

AFM images of **crystalline cell parts** show a **paramylon grain** and presumably a **lipid body**. Both serve as energy storage sites, either in form of crystalline carbohydrates or crystalline wax esters.

A large number of TEM and SEM images is available, but there have been few attempts to investigate this organism by means of AFM [4]. Our data may be compared to existing data from electron microscopy and contribute to a deeper understanding of the **correlation between structure and function.**


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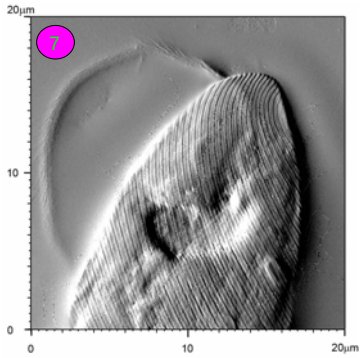

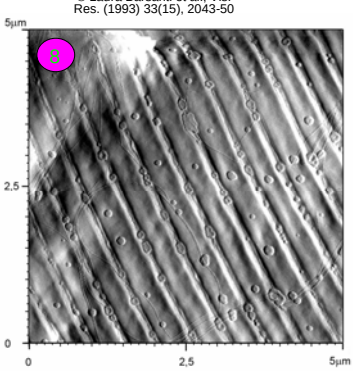

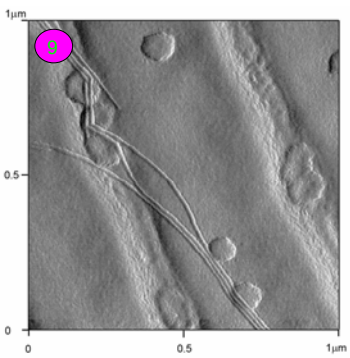

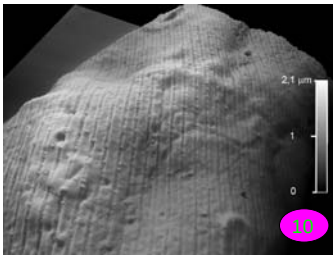

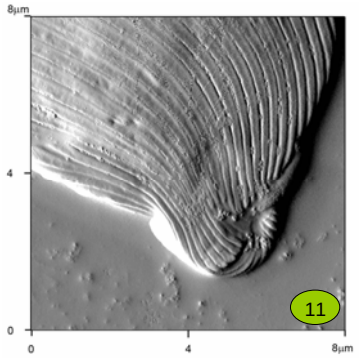

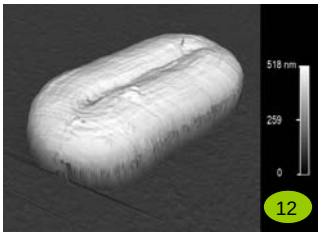

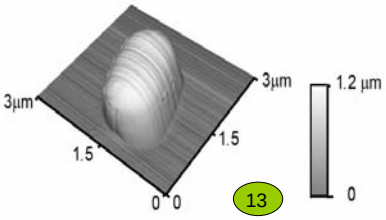

Outlook

AFM of the photoreceptor organelle is our **next goal**. The solution of crystalline parts contains only few photoreceptors, a large portion consists of lipid bodies, paramylon grains and not completely dissolved cell tissue. Therefore location of the photoreceptors is difficult and **combination with fluorescence microscopy** is needed.
As a more general outlook concerning biomaterials in material science, it is stressed that relating structure to function in biomaterials can only be the beginning of promising developments. The thermal and hydrolytic sensitivities of biological materials limit their applicability in many important synthetic material applications. **A real breakthrough requires an understanding of the basic building principles of living organisms and a study of the chemical and physical properties at the interfaces, to control the form, size and compaction of objects.**

Acknowledgements

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