

Universität Leipzig

Biophysik-Praktikum

# Forces on Cells in a Two-Beam Laser Trap (OS)



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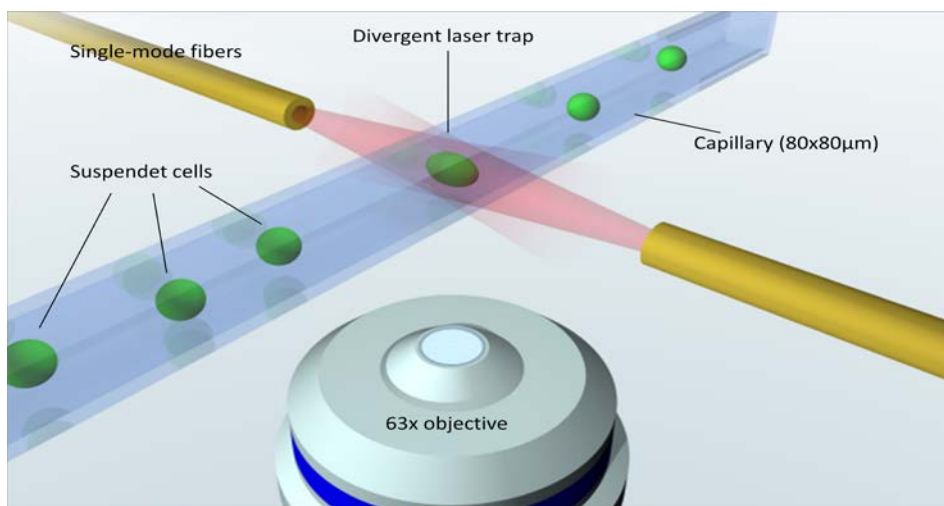
## Forces on Cells in a Two-Beam Laser Trap (OS)

### Introduction

Biological cells are the functional building blocks of life. In large structures they form tissue and organs which, taken together, make up the human body. Due to the numerous individual tasks cells have to fulfil in the body, they perform various complex activities. Many investigations have been driven by the desire to understand, predict, and influence cellular behaviour. In order to comprehend the mechanisms of biological functioning of the cellular unit it is necessary to investigate the interaction of the underlying subsystems. One of the major underlying functional systems of the cell is the cytoskeleton. It is a polymer network consisting of individual sub-networks and essential for cellular functions such as cell motility, organelle transport, mechanotransduction, and cell division. Cytoskeletal characteristics are reflected in its mechanical properties, which can be probed by rheology.

The Optical Stretcher can be used to do whole cell elasticity measurements. In contrast to other techniques the measurements can be done without touching or modifying the cell (Guck 1997). Two counter propagating divergent laser beams create an optical trap in which particles can be trapped and stretched.

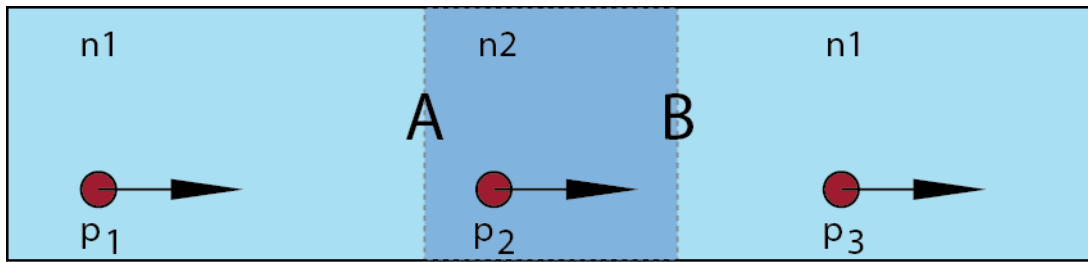
### Theoretical Background



**Figure 1: Scheme of the flow chamber of the AMOS. Cells are delivered through the flow channel and trapped and deformed in the center of the two counter propagating divergent laser beams.**

The Automated Microfluidic Optical Stretcher (AMOS, see figure 1) is a further development of the former setup advanced by a microfluidic delivery system and a computer-controlled pumping system as well as sophisticated software tools. It enables fully automated measurements of about 200 cells per hour and therefore empowers statistically significant results.

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**Figure 2:** A photon traveling in a medium of refractive index  $n_1$ , carrying a momentum of  $p_1$ . At the interface A, the photon enters another medium of refractive index  $n_2$ . At interface B, the photon exits medium 2 and enters to medium 1 again.

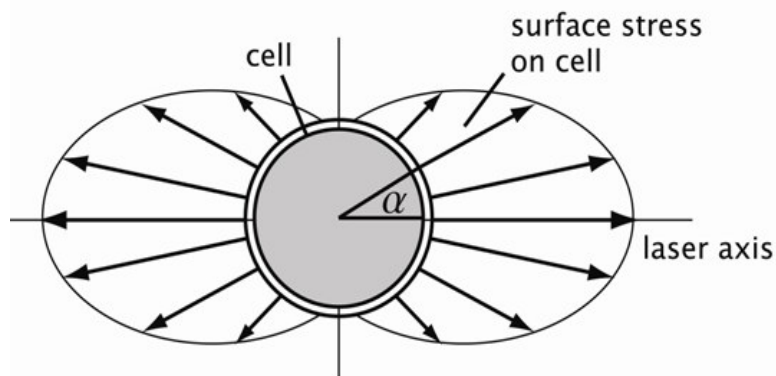
Einstein's explanation of the photoelectric effect is based on the ascription of momentum to photons. Contrary to Newton's second law, massless quasi-particles are therefore able to apply forces, according to classical transfer and conservation of momentum.

Following this explanation, a photon travelling in media of refractive index  $n_m$  carries a momentum  $\vec{p}_m$  given by:

$$\vec{p}_m = \frac{E n_m}{c_0} \vec{e}_r \quad (1)$$

where  $E = h\nu$  is the energy of a photon,  $\vec{e}_r$  is the unit vector in direction of propagation and  $c_0$  is the speed of light in vacuum.

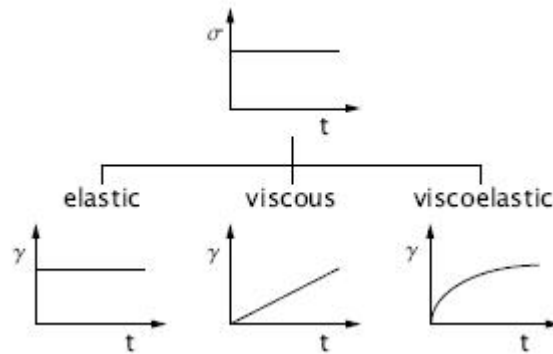
In case of a cell experiencing forces in a two beam laser trap, more complicated calculations yield a stress profile acting on the cell's surface as shown in figure 3.



**Figure 3:** Profile of the optical surface stress along the angle  $\alpha$  of a cell in a two beam laser trap (source: (Wottawah 2006)).

When a deforming force is applied to a material, it will respond either elastic, viscous or viscoelastic, an intermediate form of purely elastic or viscous behaviour, such as cells. The different responses are illustrated in figure 4.

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**Figure 4: Different responses to a constant stress  $\sigma$ . Elastic materials show an immediate response while viscous materials expand proportional to time. Viscoelasticity, as a combination of elastic and viscous behaviour, response retarded to the applied stress.**

The biomechanical behaviour of cells can be used to study cytoskeletal structures as well as diseases, known to alter cytoskeletal compositions such as cancer (Zink, Fritsch et al. 2010).

### Tasks and experimental procedures

*In experimental physics, your work is not done by creating an experiment; mostly, this is merely the beginning. After you measured "some numbers", you have to interpret them and show the significance of your measured/calculated values. In this experiment, the main focus is neither preparing an experiment nor the acquisition of data (this will be mostly done by the computer or us), but to analyse the data and to test for its significance. For this, a rigorous theoretical understanding and preparation is recommended.*

**Calculations to be done IN PREPARATION for the experiment. Results will be discussed during the Antestat.**

**!!! This will make 25% of your final mark together with the Antestat!**

Consider a photon as shown in **figure 2**, travelling in a medium with refractive index  $n_1$ . What momentum is transferred to interface  $A$  if the photon is totally reflected?

What momentum is transferred to interface  $A$  when the photon traverse interface  $A$ . What momentum is transferred to interface  $B$  when the photon traverse interface  $B$ . Compare the direction of the induced momenta at interface  $A$  and  $B$ , under the assumption of  $n_2 > n_1$ .

Given a laser power of  $P = 1.0 \text{ W}$ . Calculate the appearing forces at interface  $A$ , given  $n_1 = 1.33$  and  $n_2 = 1.37$ . Mind the relation  $\vec{F} = d\vec{p}/d\vec{t}$ . Reflections can be ignored.

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### Evaluation of two sets of data from cells stretched at different laser power.

You will get raw data from cells stretched at 800 and 1200 mW showing the diameter of the cells along the laser axis in time. Choose an appropriate data processing program (e.g. Origin, MatLab, Mathematica) for data evaluation.

Determine the relative deformation of the cell with time. Use initial diameter of the cells during trap for normalisation.

The maximum deformation at the end of stretch is of special interest. Plot histograms of this value for both data sets. Determine mean value and standard deviation.

Do the histograms resemble a Gaussian distribution? Use statistical tools to verify your answer. Bonus: Which probability distribution would eventually better describe your data? Show with fit of distribution curves the better matching.

How much do the mean values of the two measurements differ from each other? Is this change significant? Use statistical tools (e.g. Student's t-test, Chi-square-test ...) and give significance level. What are the limitations of your chosen significance test?

What is the impact of laser power on the acting forces?

Plot mean values of deformation over time for the data sets of the two measured laser powers. Does the difference mirrors your expectation of the influence of the laser power on the applied force?

Is there a change in deformation behavior (shape of deformation curve)? *Bonus: Do cells react more viscous or more elastic at higher forces?*

## References

Guck, J. (1997). The optical stretcher, a novel, noninvasive tool to manipulate biological materials. Department of Physics. Austin, TX, University of Texas at Austin.

Wottawah, F. (2006). Optical Cell Rheology: From the Microscopic Origins of Cellular Elasticity to Oral Cancer Diagnosis. Institute of Soft Matter Physics. Leipzig, University of Leipzig: 118.

Zink, M., A. Fritsch, et al. (2010). "Probing the physics of tumor cells from mechanical perspectives." Cell News - Newsletter of the German Society for Biology **36**(4/2010): 17-21.