

Introduction

Microfluidic bioreactors offer a huge potential for in-vitro testing of long-term toxicity in order to ultimately replace the use of animals. To this end, cultured cells are challenged with repeated doses of a chemical compound, while their responses are monitored. Our project's aim is a human liver-simulating microdevice mimicking the liver structurally and functionally over a period of one month. Continuously controlling culture conditions is indispensable for maintaining a cell culture for such a long time. Key parameters include the glucose and oxygen concentration as well as the pH value. Measuring these parameters provides not only a means of quality control, but also yields input data for a planned automated feedback control of the microreactor. However, sensing in a microfluidic environment poses other requirements than on the macroscale. Main challenges comprise miniaturization as well as desired lifetime. Hence, commercial sensors need to be adapted and novel sensors have to be developed in order to achieve this aim.



Cell culture condition monitoring in microfluidic bioreactors

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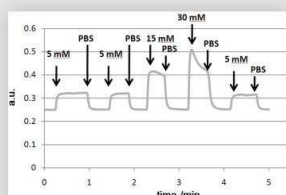
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Conclusion

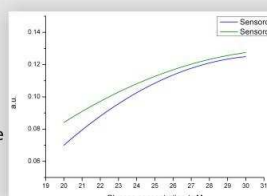
First steps were accomplished towards adapting a commercial blood glucose sensor to a microfluidic cell culture reactor. As proper functioning of the sensor chip in our measurement setup was proven, tests with complex fluids, such as conditioned culture media, are scheduled next. Subsequently to this evaluation process, the sensors will then be integrated into the reactor. The optical oxygen sensing technique presented here was shown to be stable and reliable in the microfluidic environment. Hence, the main objective is now on further integration in order to harness the stability and precision provided by this sensor in the planned bioreactor.

Glucose Detection

Results



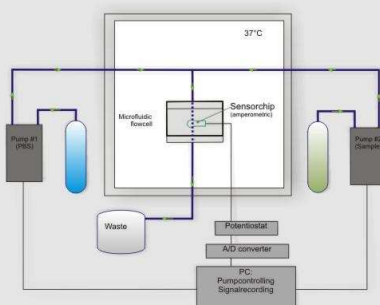
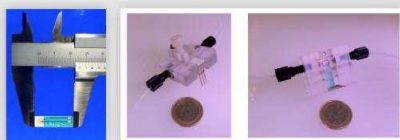
Exemplary measurement of three different glucose concentrations in phosphate buffered saline (PBS). The sensor's output is excellently reproducible and has a high signal-to-noise ratio.



Calibration curves of two different sensor chips. The glucose concentration in cell culture medium is relatively high (20 - 30 mM). In this range, the output signal is no longer linear, necessitating calibration.

Method

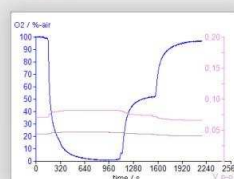
Amperometric sensor chip (BST, Berlin, Germany) on a ceramic substrate employed in a microfluidic flowcell with an inner volume of only 2 µl.



Fully automated microfluidic setup driven by a computer. The sensor chip is alternately flushed with sample and PBS. The raw signal (current) is converted by an A/D converter and subsequently processed and recorded by the computer. The sensor chip is placed in an incubator at 37 °C in order to simulate the environment of the future bioreactor.

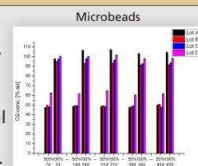
Oxygen Detection

Results

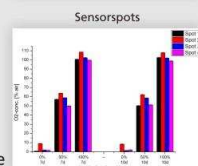


Measurement with calibrated oxygen sensor. Blue curve: oxygen concentration, pink and brown curves: signal amplitude. The 100% value refers to the atmospheric oxygen concentration (21 vol.-%).

Signal recorded over 6 weeks for assessing stability. Except for a slight initial shift, the values remain constant.

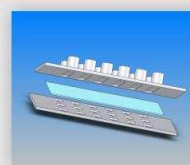


Same as above, but for sensor spots. Measurement over 10 days indicates stable and reproducible behaviour.



Method

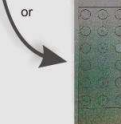
Home-made flowcell used for simulation of the fluidic environment of the future bioreactor.



middle well equipped with:



Phosphorescent polystyrene microbeads (Ø 50 µm).

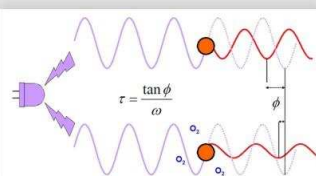


Thin-film spots carrying the phosphorescent dye.



Reading out a sensor spot.

in cooperation with
ColibriPhotonics
and
Fraunhofer IAP



This optical measurement is based on luminescence quenching. The energy transfer from the excited dye molecule to oxygen leads to a decreased phosphorescence lifetime. Hence, a phase shift between excitation and emission light is induced and detected. The lifetime decrease correlates with the oxygen concentration.

