Microbial biosensor for industrial online analysis

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The role of microbes in biotechnological production processes is many-faceted, from being the microscopic workhorses of the biochemical process applied, to the saboteurs invading the production line and spoiling the entire yield. Beneficial or detrimental, these little organisms are difficult to track down, control, and tame. State-of-the-art techniques for their detection are laborious with typical several days of cell culturing, followed by some kind of microbiological testing on their nature. While such approach eventually gives a precise taxonomy of the species under test, it is quite obvious that it will be of little use as a permanent in-field process monitor.

Here, a novel approach to label-free bacterial and fungal detection is presented. It is based on fluorescent, specifically functionalized polymer microbeads, which bind to their target organism while freely floating in the analyte. In contrast to state-of-the-art label free biosensors, which are mostly embodied as microfluidic devices with the bio-functionalized surface formed on one of the channel walls, the microbeads cruise through the analyte and thus raise the chance of hitting their specific targets. Sensor read-out is performed remotely by optical means, i.e., fluorescent excitation of the organic dye embedded into the polymer beads and recording of their fluorescent emission (Fig.1 left). A numeric algorithm based on Mie theory is then applied to the resultant fluorescence spectra for the assessment of potential binding events at the sensor surface (**F**luorescent **R**esonator **S**ignature, FRS). Since the sensors are microscopic particles, their number can be made sufficiently high to allow for reliable statistics on the concentration of the targeted microbe in the monitored bioreactor system.

Figure 1 shows a schematic of a basic implementation of an FSR sensor. Sensor beads are added to a reactor line containing the analyte to be screened for microbes. To avoid contamination of the production medium with sensor beads, the effective detection line can bypass the main line. The microbeads, freely floating with the analyte, are interrogated optically at a suitable location of the detection line, thereby generating some fluorescent resonator signature (FRS) inside of the microbeads passing the spot of detection. Depending on the particles' specific surface functionalization, only selected microbes bind to the microbeads, thereby causing changes in the FRS of the respective sensor particle (Fig. 1 right). This sensor scheme can be easily expanded to the detection of more than a single kind of microbe by variation in the specific bio-functionalization of the microbeads applied.

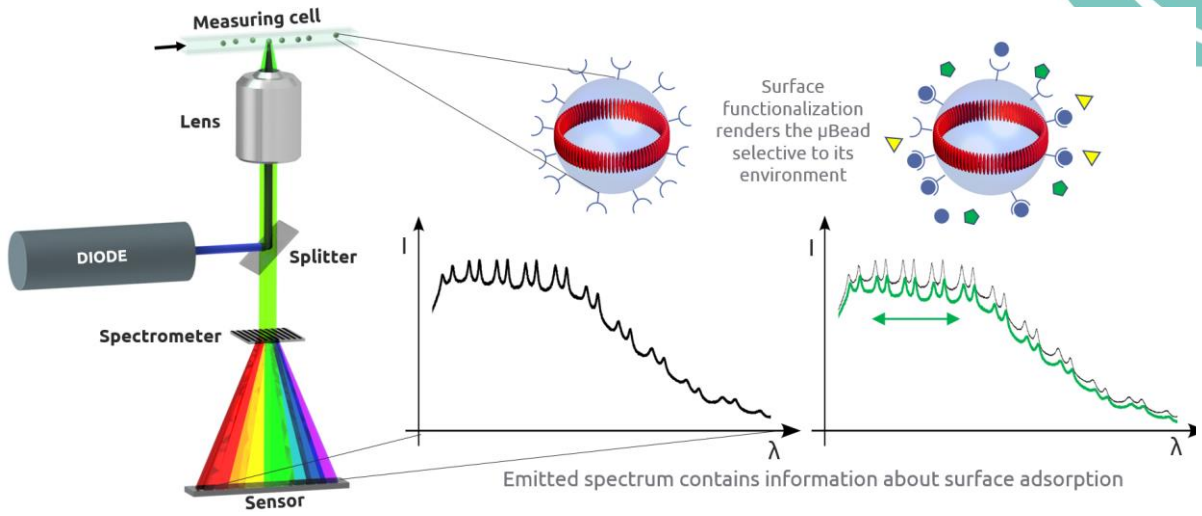

Fig. 1: Schematic of Fluorescent Resonator Signature (FRS) Sensing: (left) optical set-up for excitation and read-out of FRS of the freely floating sensor beads; (right) illustration of the change in FRS by specific binding of some targeted microbe to the microbead surface.