**Measurement of bacterial biosensor activity at the single cell level: outlooks to better understand the availability of pollutants at microscale**

Robin Tecon, Jan Roelof van der Meer, DMF, UNIL

Bacterial biosensors (also called whole-cell living bioreporters) are micro-organisms engineered to detect target chemical compounds via the production of measurable reporter proteins. For more than one decade, this technology has proven its usefulness in monitoring a wide variety of pollutants, including heavy metals, halogenated aromatics and alkanes. Here we work with the biosensor Burkholderia sp. RP037, which produces GFP as a reporter signal when it is actively degrading phenanthrene - an organic contaminant which is a common component of crude oil. Phenanthrene is poorly bioavailable for bacteria in the environment, because of its low water solubility and its high tendency to sorb to the organic phase in soils and sediments. The result of this is that the rate of phenanthrene degradation in natural environment is limited by physicochemical parameters (e.g., solubility and diffusivity of the chemical, distance between pollution source and degrader) and not by the physiological or genetic properties of the bacteria degrading phenanthrene. In order to investigate the physicochemical factors influencing bioavailability of phenanthrene for Burkholderia, single cell analysis is advantageous. We have used epifluorescence microscopy and scanning laser confocal microscopy, together with specific fluorescent tagging of Burkholderia sp. RP037, to visualize and detect bacteria and determine their phenanthrene degradation activity. Quantification of single cell GFP production in Burkholderia cells has permitted calibration of the phenanthrene flux to the cells. This will be extended by double marked strains and the use of miniaturize flow chambers in order to measure the phenanthrene flux from different contaminated materials.

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