

### **Abstract for the general public**

Droplet microfluidics has been one of the most dynamically developing branches of analytical chemistry in the last 20 years. One of the main goals of research in microfluidics is the development of analytical systems (so-called Lab-On-a-Chip) for various applications in experimental biology, medical diagnostics or materials chemistry. Droplet microfluidic systems allow the formation of thousands or millions of microscopic droplets of equal volume in the pico- and nanoliter range. The ability to create such a large number of droplet microreactors has allowed for many spectacular successes of this method in recent years - e.g., genomic profiling of single cells, detection of single DNA molecules, or measurement of the activity of millions of variants of enzymes or antibodies. Researchers are currently able to create vast numbers of drops, but in most applications, these drops have the same chemical composition, which significantly limits the spectrum of their applications and further development of this method. This problem has been solved to some extent by controlling the position of the drops (e.g. their order in the tube) or by using mixtures of optical markers (e.g. fluorescent dyes). This way, it was possible to "encode" the chemical composition of hundreds or several thousand droplets in one experiment. However, this does not cover the full potential of the method, which allows the creation of hundreds of millions of droplet bioreactors during an experiment lasting several hours.

In this project, we will address the challenge of limited encoding capacity by using mixtures of DNA fragments that will encode the chemical composition of each microdroplet. Deciphering this composition after the completion of a reaction will be possible by uniquely labeling these fragments in each of the million droplets and using high-throughput next-generation sequencing (NGS) technologies to read DNA sequences. In this way, we will synergistically combine the enormous throughput of droplet microfluidics and NGS technologies and remove one of the main limitations in ultra-high-throughput screening with Lab-On-a-Chip systems.

To demonstrate the unique capabilities of the new method, we will study the consortia of environmental bacteria to select more efficient cellulose-degrading strains and antibiotic-producing microorganisms. The very high throughput of the method combined with read-out using DNA sequencing will also allow us to determine the structure of microbial populations and examine interactions between bacterial strains depending on the availability of nutrients in microcultures.

We anticipate that the newly developed method will find many future applications in various high-throughput microbiology studies. The technique will enable studies towards a better understanding of the physiology of bacterial consortia from different environments, including potential biotherapeutic strains from the gut microbiome. Very high throughput will allow for the optimization of microbiological media in order to grow as many environmental strains as possible and to study the interactions between antibiotics. The method may also find applications in other areas of experimental biology, e.g. to optimize media used in the differentiation of mammalian cells or the formation of organoids.