Introducing high pressure microfluidic tools to study extreme environments

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Over the last 10 years, a new field of investigation called "high pressure microfluidics" has gained increasing interest [1]. It combines the advantages of microfluidics (*i.e.* size reduction, fast screening, *in situ* analyses, high reproducibility, hydrodynamic control, improvement of heat and mass transfers, *etc.*) with fluid systems used under high-pressure (and high temperature) conditions. The development of high-pressure microfluidic has opened up many opportunities to study and characterize a large number of processes using high-pressure fluids such as hydro- and solvothermal processes, thermodynamics measurements [2], studies of geofluids flows in model porous media [3] or supercritical fluids [4]. More recently, it appears that such tools are particularly well adapted to investigate microbes living within extreme environmental conditions such as deep biosphere microorganisms.

A majority of Earth's prokaryotes resides in the deep biosphere (deep-sea [5] or deep underground environments) where little is known about how inherent elevated pressures impact the underground biogeochemistry and the inhabiting microbial communities. The deep biosphere represents the unseen majority of the total biosphere on Earth. Currently, the complex sampling procedures on deep sites with specific samplers result in small amounts of biological materials, while the sample analysis mostly requires decompression, which induces uncontrolled biases during the investigations. Although some HP experimental means exists, microbiology under extreme conditions is still scarcely studied. Indeed, conventional cultivation and analysis techniques offer limited *in situ* characterization thus narrowing the ability to investigate deep subsurface microbial communities. HP microfluidics tools are able to overcome these limitations being able to propose fast screening approaches and *in situ* monitoring in real conditions, both in batch and continuous mode using small quantities of biological materials.

In this presentation, we will detail first the interest of this technology and the different strategies developed to manufacture and use high-pressure microreactors. Then, we will present the use of on-chip biocompatible high-pressure geological laboratories for the culture and the study of methanogenic microorganisms living in deep geological environments. As part of the "BIG MAC" ERC project [6], which aims at mimicking deep geological environments, we study the bioconversion reactions of CO_2 into methane by methanogenic archaea. These tools could provide new insights into bioremediation process to restore CO_2 as a valuable energy resource (*i.e*. CH_4 *via* methanogenesis process) and could find wider applications in geological-related. Eventually, we will emphasize on the use of these tools to wider applications on deep-sea field studies with the example of an oceanographic cruise to sample and analyze deep-sea vent fluids.

In conclusion, high-pressure microreactors turn out to be excellent experimental tools for investigating microbiology under extreme conditions at the lab scale. After summarizing the advantages of these new experimental approaches compared to traditional instrumentation, we will give some perspectives concerning other future applications of this technology applied to the study and use of extremophile microorganisms (*e.g*. thermophile, piezophile) for various engineering processes aiming at addressing environmental issues.

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