DEVELOPMENT OF A NOVEL AUTOMATED FLOW-NMR PLATFORM BASED ON SEGMENTED-FLOW ANALYSIS SAMPLE TRANSFER

Bert Wouters[a], Paul Miggiels[a], Roland Bezemer[b], Elwin A.W. van der Cruijsen[b], John Gauvin[b], Klaartje Houben[b], Paul Zuijdwijk[b], Amy Harms[a], Adriana Carvalho de Souza[b], and Thomas Hankemeier[a*]

[a]Analytical Biosciences and Metabolomics, Systems Biomedicine and Pharmacology, Leiden Academic Centre for Drug Research, Leiden University, Einsteinweg 55, 2333 CC Leiden, The Netherlands

[b]DSM Biotechnology Center, Alexander Fleminglaan 1, 2613 AX Delft, The Netherlands

It is becoming increasingly important to analyze vast numbers of samples in fields such as industrial biotechnology and life sciences. Nuclear magnetic resonance (NMR) spectroscopy has proven itself as a powerful analytical technique capable of providing quantitative molecular information of complex samples in a cost- and time-efficient manner. For rapid screening of samples, fast and robust sample transfer is crucial to increase the sample throughput. Segmented-flow analysis NMR (SFA-NMR) with fluorinated oil was recently introduced for sample transfer, employing a biphasic flow consisting of samples isolated by oil segments. It has numerous benefits over existing flow-NMR methods (direct-injection, DI-NMR, flow-injection analysis, FIA-NMR, and segmented-flow analysis with air bubbles to separate samples), most importantly lower sample consumption, zero sample dispersion, and low carryover. However, SFA-NMR with fluorinated oil has only been reported for microcoil-NMR, or for commercial glass flow cells that are modified with an internal fluorosilane coating.

We have developed a novel polychlorotrifluoroethylene (PCTFE) fluoropolymer flow cell combined with segmented-flow analysis for an automated flow-NMR screening platform based on a conventional 5.0-mm NMR probe. An in-house developed Python interface was used to orchestrate the sample injection, transfer, and acquisition, using the Bruker HyStar protocol for communication with TopSpin at the NMR. The developed flow cell can be used with a standard CryoFIT accessory and a 5.0-mm NMR cryoprobe and features reversible connections pressure-resistant up to 1.5 MPa. 1D NMR spectra were acquired under full automation from 96-deepwell plates with a sample-transfer time to the flow cell of 42 seconds. Sample-to-sample carryover for aqueous standards such as sucrose, maleic acid, and citrate was in the 0.4-0.6 % range. Finally, the applicability of the system was demonstrated by the screening of complex multi-component samples such as non-alcoholic wine.

Keywords: Metabolomics; biphasic flow; high-throughput; PCTFE