New Approaches to On-line and *In-Situ* NMR Reaction Monitoring:Fast and Furious Data

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There are multiple applications of nuclear magnetic resonance (NMR) spectroscopy across a variety of industries. This powerful analytical technique has been used to study the structure of molecules, and the interactions between them for the past 60 years. Its broad range of uses in the pharmaceutical, biomedical, food science and many other industries make NMR an invaluable tool.

The focus of this paper is a relatively new capability of NMR for monitoring reactions in real-time.[1] Two purpose-designed flow tubes are described, and experimental data is presented. The flow tubes are offered as standard accessories for Bruker spectrometers that operate in the range from 300 to 950 MHz equipped with a 5 mm probe.[2]

- The InsightMR flow unit is designed to circulate reacting materials from an external reactor through the magnet for rapid spectral acquisition, thereby enabling on-line monitoring.
- A complementary design, designated InsightXpress,[3,4] can be used for *in-situ* measurements where reagents are mixed directly inside the magnet. This enables measurement of the kinetics of very fast reactions.

These two solutions have been designed for multiple end uses including: measuring reaction kinetics; understanding reaction mechanisms; maximising chemical yields, and determining the effect of process variables (e.g. stoichiometry, temperature etc.) on the reaction under study. As shown by several papers and publications, NMR is capable of achieving such insights and the systems described here are the first purpose-designed flow tubes that can help achieve these important goals.

General Description

In the past, the challenges facing this range of applications within the industry included the fact that there was a lack of appropriate hardware to track reactions under realistic conditions. Also, there was a lack of software to process the data properly, and present it appropriately.

In this article, we discuss solutions to these challenges in the form of integrated hardware and software platforms (InsightMR and InsightXpress) which are set to revolutionise on-line and *in-situ* reaction monitoring. Further, these platforms are entirely compatible with the standard laboratory based NMR infrastructure from Bruker and therefore data, together with associated insights derived from multiple approaches (on-line, *in-situ* and at-line) can be easily merged.

Comparison of At-line, On-line and *In-Situ* Monitoring[5]

The long-established at-line approach to analysing a chemical reaction with NMR has been to extract a sample (or multiple samples) from the reaction under study, for subsequent analysis in a central analytical laboratory. This is an intrusive and labour intensive practice. In addition, at-line has the important disadvantage of being slow, with cycle times (i.e. from the reaction actually occurring, to results being presented) being in the range of tens of minutes.

In contrast, the on-line configuration from Bruker uses the InsightMR flow tube and samples the reaction directly from a reaction vessel, then circulates the sample through the magnet system. This on-line approach is carried out in a controlled manner, so that temperature and pressure are not affected (Figure 1). The time delay for on-line monitoring using such a system is typically less than one minute.

In a more recent development, *in-situ* monitoring is made possible through the use of the InsightXpress unit (Figure 2). In this configuration, reactants are mixed inside the magnet and quickly pumped into the NMR probe through a capillary with a delay of only 130 milliseconds. (Note that this a remarkably short time period, and is a key aspect of the performance of the InsightXpress flow tube). Overall, insightXpress enables very fast reactions (with half-lives as short as a second) to be tracked via NMR spectroscopy.

Figure 1: On-line reaction monitoring with InsightMR

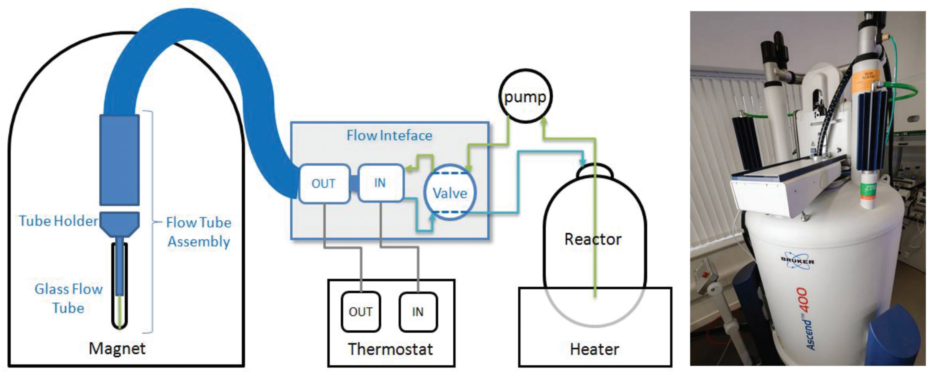
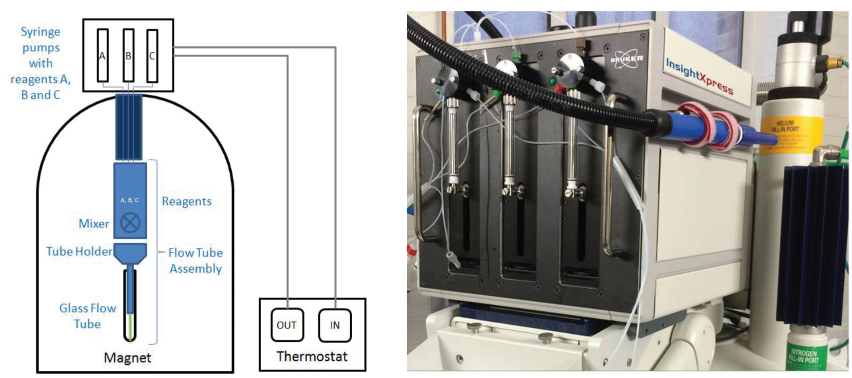


Figure 2: In-situ reaction monitoring with InsightXpress



On-line Reaction Monitoring

Benefits

A key advantage of on-line reaction monitoring is the ability to collect compositional information and derive kinetic data from chemical processes in real laboratory conditions (stirring, temperature, pressure) and also close to the beginning of a reaction. This capability is important, for example, when attempting to observe early-forming intermediates in a reaction. This approach also allows simultaneous monitoring (via the reaction vessel) with other analytical techniques, such as MS, IR, UV and pH, and enables scientists to accumulate comparable sets of data from these additional techniques.

Case Study: InsightMR

Figure 3 describes the integrated use of NMR and infrared (IR) spectroscopy to improve understanding of the synthesis of a precursor to an active pharmaceutical ingredient (API).[5] Identification of reaction intermediates (in this example, pivalic anhydride) is key to comprehensive chemical reaction understanding and therefore vital to reducing costs and time when manufacturing APIs.

In-situ Fast Reaction Monitoring

Benefits

Another difficulty often faced during reaction analysis is speed. Extremely fast reactions can be challenging to monitor and study; therefore understanding and control of these reactions is limited. Using the InsightXpress unit described here, data from fast reactions can be captured and monitored in order to identify optimum conditions and develop improved understanding. Automated delivery and mixing of reagents at different ratios and a post-reaction solvent flush (wash) makes this system ideal for rapid, streamlined screening of reaction condition, a key part of process optimisation and scale-up. Additionally, accurate temperature control during experiments, and a high degree of reproducibility are further advantages of this approach.

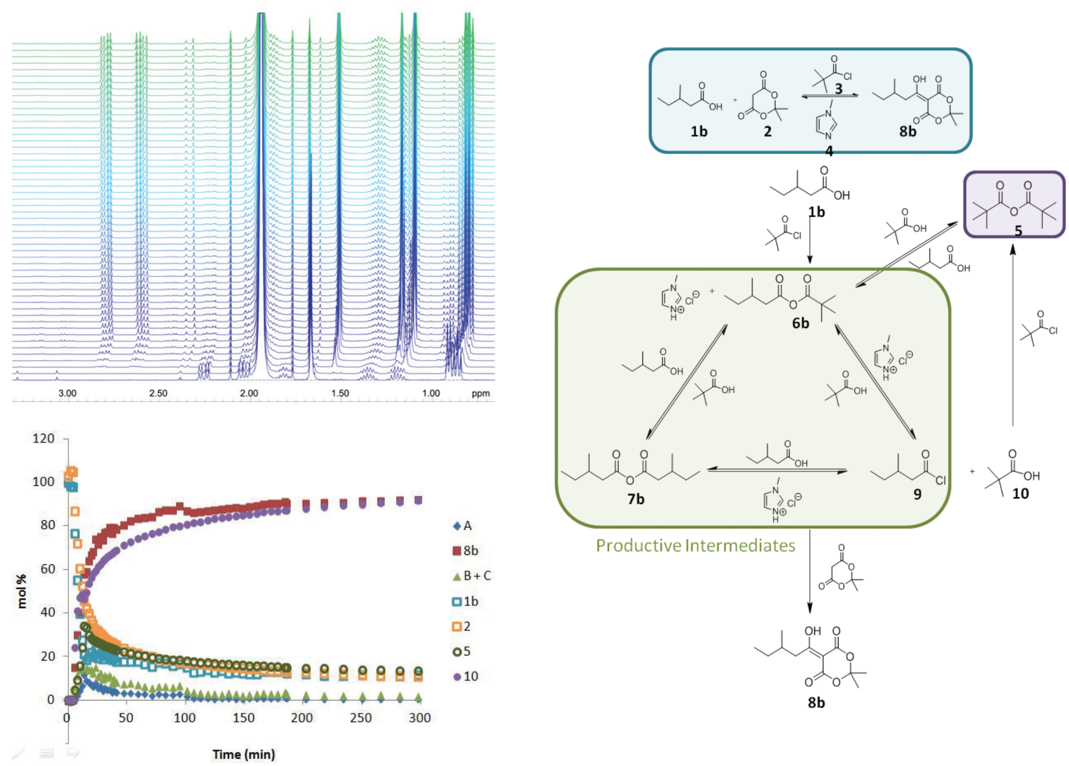
Case Study: InsightXpress

A second case study (Figure 4), explains how stop-flow (SF) techniques can be used to evaluate very fast reactions, and therefore find optimum reaction conditions. In this case, SF-NMR and SF-IR worked in tandem to facilitate complete

CASE STUDY: *InsightMR. In this example, on-line NMR is able to elucidate intermediate structures, which can in turn be associated with the IR trends. Therefore NMR complements IR and greatly enhances reaction understanding.*

One of the many applications of NMR spectroscopy is the characterisation of reaction intermediates in order to improve the understanding of the synthesis of an active pharmaceutical ingredient (API). Through this method it is possible to quantitatively profile every material involved in the reaction, and provide insights into the reaction mechanisms. This case study describes the course of a reaction to form Meldrum's acid adduct, the first step in the production of an API.

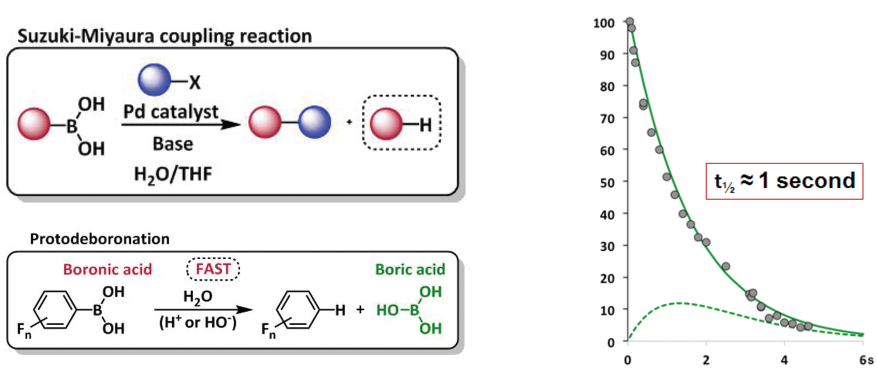
Figure 3



CASE STUDY: *InsightXpress: Very fast reactions can be tracked by NMR spectroscopy*

The Suzuki-Miyaura cross-coupling reaction is an example of a very fast reaction which presents a monitoring challenge. Effective cross-coupling of poly-fluorophenyl boronic acid with an eletrophile must occur under optimum conditions; otherwise boronic acids can protodeboronate which reduces yields and cross-coupling efficiencies. Stop-flow mechanisms can be used to study the protodeboronation reaction, allowing gaining insights into the reaction mechanisms and ultimately increasing the yield of the Suzuki-Miyaura reaction.

Figure 4



understanding of rapid reactions, as exemplified by this cross-coupling reaction (Suzuki-Miyaura proto-deboronation side reaction).[6] It is important to note that this approach of using the InsightXpress also enables improvements in productivity: in this particular example, manual sample preparation and handling would be possible but would require one to two week's work compared to approximately three hours with *in-situ* SF monitoring using insightXpress flow unit.

Solving the Integration Problem

The case studies described herein (Figures 3 and 4) give a clear picture of the benefits of utilising on-line or *in-situ* NMR to study chemical reactions. The flow units described are fully integrated with the standard Bruker infrastructure (both hardware and software) and can overcome some of the key challenges that have faced the industry. Further, we have shown an example of how these technologies can be used in combination with another analytical technique (IR): a combination that can be very useful.

Conclusions

NMR spectroscopy has been utilised in analytical laboratories for decades; assisting industrial and academic scientists in understanding the mechanisms behind a variety of reactions.

Monitoring by NMR facilitates process understanding and optimization, leading to improved yields and reduced costs. Previously, only at-line methods of NMR reaction monitoring were possible but this had the important disadvantage of not enabling the early phase(s) of the reaction to be observed (and therefore important insights could be missed). Also, at-line experiments can cause too much disturbance to the physical conditions of the reaction (i.e. temperature and pressure).[7] Employing the flow tubes described in this note enables both these major disadvantages, and several others, to be overcome. The new platforms consist of a flow unit, which achieves tight temperature control via its transfer lines, whilst the associated software allows interactive data analysis. Together, this facilitates the accumulation of quantitative data in real-time, in a very practical manner.

References

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[5] These terms are consistent with the definitions contained in the document "Guidance for Industry PAT – A framework for Innovative Pharmaceutical Development, Manufacture and Quality Assurance". Food and Drug Administration, 2004
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