Journal of Labelled Compounds and Radiopharmaceuticals

Published online 13 May 2010 in Wiley Interscience (www.interscience.wiley.com) DOI: 10.1002/jlcr.1761

10th International Symposium on the Synthesis and Applications of Isotopes and Isotopically Labelled Compounds—Professor John R. Jones Memorial Lectures, Monday, June 15, 2009

SESSION CHAIR: DAVID HESK

Schering-Plough, USA

Abstract: The first Session of the Conference was dedicated to the memory of Professor John R. Jones. Labelling with Deuterium and Tritium was the common theme. William J.S. Lockley, Frank Tang and Shuiyu Lu gave presentations.

Keywords: Tritium; Deuterium; Catalysis; Hydrogen Exchange Reaction; Professor John R. Jones; fluorine-18 chemistry; micro-reactors

CATALYTIC HYDROGEN ISOTOPE CHEMISTRY: 25 YEARS OF COLLABORATION WITH JOHN R. JONES

WILLIAM J. S. LOCKLEY

Division of Chemistry, Faculty of Health and Medical Sciences, University of Surrey, Guildford, GU2 7XH, UK

Abstract: The long-term collaboration with Prof. John R Jones has enabled the development or discovery of a range of one-step ^{3}H- and ^{2}H-labelling approaches. These include; (a) $RhCl_3.3H_2O$ and COD.Ir.Acac catalysed *ortho*-exchange of aromatic acids, amides, benzylamines, anilides, sulphonamides, etc, using isotopic water as the donor in DMF or DMA solution, (b) the COD.Ir.F_6Acac catalysed *ortho*-labelling of anilines, benzylamines and other *N*-heterocyclics using isotopic hydrogen gas in DMA at RT, (c) the α- and β-labelling of piperidines, piperazines and other secondary amines using catalytic $Ru(CO)_6Cl_2$ or $(C_6H_5)_2RuCl_2$ with deuterium oxide in DMSO, (d) the α-labelling of pyridines and other *N*-heteroaromatics using rhodium and ruthenium heterogeneous catalysts with isotopic hydrogen gas in THF at RT and (e) the preparation of an efficient and clean polystyrene-based Ir(I) phosphine catalyst for the labelling of aromatic esters, aromatic ketones, sulphones, amides, anilides, etc, using isotopic hydrogen gas in dichloromethane at RT. Latterly, screening for the terminal-methylene labelling of 1-monosubstituted and 1,1-disubstituted alkenes has provided new catalytic systems for this transformation. In addition, it has provided evidence for the significance of secondary metal alkyl intermediates in the general mechanism of catalytic alkene exchange and reduction. These studies were facilitated by the application of a ^{3}H-NMR cryoprobe.

Keywords: isotope-exchange; *ortho*-labelling; catalysis; alkene-exchange; hydrogenation-mechanism; deuteration; tritiation

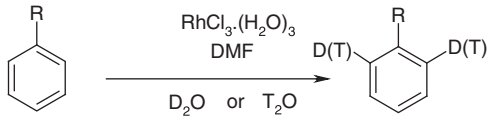
Introduction: Professor John Richards Jones was a very significant figure in the field of isotopic chemistry. His collaborations were many and varied, and his influence was very extensive. This paper describes a range of studies either carried out in conjunction with, or which received some useful input from, Prof. Jones during the 25 years of our collaboration. It describes useful labelling procedures and mechanistic investigations which have broad applications or significance for our field of science. It also gives a necessarily brief resume of the further developments which have been achieved in some of these fields since his retirement and death.

For reasons of brevity other areas of our collaboration; ^{3}H-NMR cryoprobe development,[1a] dehalogenation studies,[1b,j] iodination methods,[1c] microwave applications,[1d] ligand receptor interactions,[1e] isotopic data processing applications[1f,g,h] and much of the work on alkene and alkyne reduction and exchange[1i,j] have been omitted, but are referenced above for those with interests in these areas. The paper therefore concentrates solely upon the following two areas.

Results and Discussion: *Part 1. Collaboration with AstraZeneca on tritium-labelling methods.*

The initial aim of the AstraZeneca (AZ)/Surrey collaboration was to produce general and practical one-step isotope exchange procedures for labelling polar xenobiotics with tritium. The project was initiated by a thorough study of the $RhCl_3.3H_2O$-catalysed

ortho-labelling approach (Figure 1) already discovered at AZ[2a,b,c] in the early 1980s and subsequently utilised in the preparation of ^{3}H-labelled anti-allergy pharmaceuticals at high specific activity.[2d,e]



R=CO_2H, $CONH_2$, CONHR', NHCOR', CH_2NH_2, etc.

Figure 1. $RhCl_3$-$3H_2O$ catalysed ortho-exchange.

The Surrey studies led to the discovery of an interesting, more stable, though more limited, catalyst in ruthenium acetylacetonate,[2f] as well as to a better appreciation of the kinetics[2g] and utility[2h] of such novel metal-catalysed *ortho*-labelling systems.

The application of parallel catalyst screening at AZ[3a,b] subsequently identified a new highly active catalyst, **1a**, (Figure 2) which was more active for most substrates, more stable and could be used at lower temperature or under microwave irradiation. Subsequent optimisation of the structure led to a further even more active catalyst, **1b**, (Figure 2) which, in addition to retaining the $RhCl_3.3H_2O$-like activity with isotopic water,[3b] could also be utilised with an isotopic hydrogen gas donor, at RT in DMF or DMA, for the *ortho*-labelling of anilines, benzylamines and some *N*-heterocyclics.[3c] Moreover, since the functional group selectivity of this catalytic system depends upon the isotope donor used, labelling orientation can be controlled for appropriately functionalised molecules.[3d]

Subsequent screening/optimisation campaigns at Surrey then led to the identification of several effective Rh & Ru heterogeneous catalysts for the α-labelling of pyridines, pyrazines, quinolines, isoquinolines and other such *N*-heterocyclics using isotopic hydrogen gas in THF at RT.[4a] Recent reports[4b,c] have shown that, in some cases, these catalysts can be even more effective when used in conjunction with additives such as the Crabtree catalyst.

A similar screening approach also yielded two new catalysts, **2** and **3**, (Figure 2) for the α- and α,β-labelling of piperidines, piperazines, and other secondary amines using an isotopic water donor in DMSO.[5a] These catalysts promise to be even more effective than the alternative $(PPh_3)_3RuCl_2$ catalyst which has already been extensively utilised in the rapid tritium labelling of pharmaceutical agents.[5b]

Lastly, to facilitate the rapid tritium labelling approach to drug discovery developed at AZ, a highly-active solid-phase analogue, **4**, (Figure 2) of the homogeneous iridium systems widely-utilised for ^{3}H-labelling was developed[6a] and shown to lead to a much cleaner labelling procedure[6b] than the standard homogeneous catalysts.

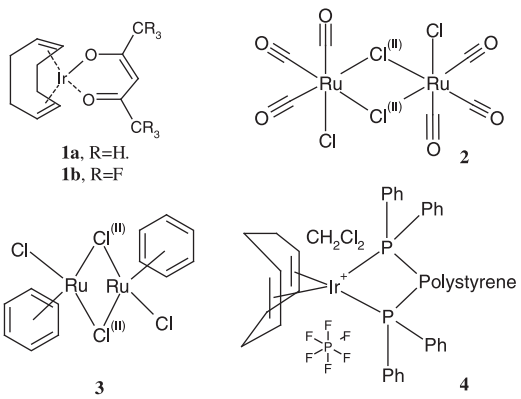


Figure 2. Structure of some catalysts for one-step exchange-labelling.

Overall therefore, the collaboration facilitated the development of catalytic one-step ^{3}H- and ^{2}H-labelling procedures for compounds containing a wide variety of common functional groups. Moreover, all the above catalysts are either commercially available or are easily prepared, without specialist apparatus in a single step, from commercially available precursors.

Part 2. Collaboration with the ATHENA consortium on alkene reduction and exchange mechanisms.

Following a productive national collaboration on catalytic mechanisms between industry and a number of universities[7] a wider international consortium was constituted under the acronym ATHENA[8] to understand and improve important catalytic processes. Surrey was involved in providing isotopic support for studies of alkene and alkyne hydrogenation. In particular many ^{2}H- and ^{3}H-NMR studies of the isotopic exchanges taking place during the hydrogenation of a wide selection of alkenes and alkynes with DT gas were carried out.[9,1i] Only the results of studies with terminal alkenes are reported here.

The incomplete hydrogenation of terminal alkenes with D_2 gas over Pd, Ru, and Ir heterogeneous catalysts yielded a mixture of **8, 9, 10** and **11** (Figure 3), together with the original unlabelled alkene. Such mixtures proved particularly mechanistically informative.

Journal of
Labelled Compounds and
Radiopharmaceuticals

They showed a clear preference for exchange and hydrogenation routes which involved secondary rather than primary metal alkyls. Moreover the degree of exchange of the *cis* and *trans* terminal protons of the exchanged alkene were equal in extent even at early time points and at low percentage reaction, suggesting the complete loss of stereochemical integrity for these protons during the exchange/hydrogenation process. This is consistent with the ability of the alkene to present either face to the catalytic surface and with the facile bond rotation available to subsequent surface species such as **6** and **7** (Figure 3).

Overall, the catalyst screening results from terminal alkene studies were supportive of modern variants of the Horiuti-Polanyi hydrogenation mechanism[10] (E.g. Figure 3) involving surface alkyls[11] such as **6** and **7,** but is also consistent with the intermediacy of di-σ-bonded species[12] such as **5** which can equilibrate with **6** and **7**.

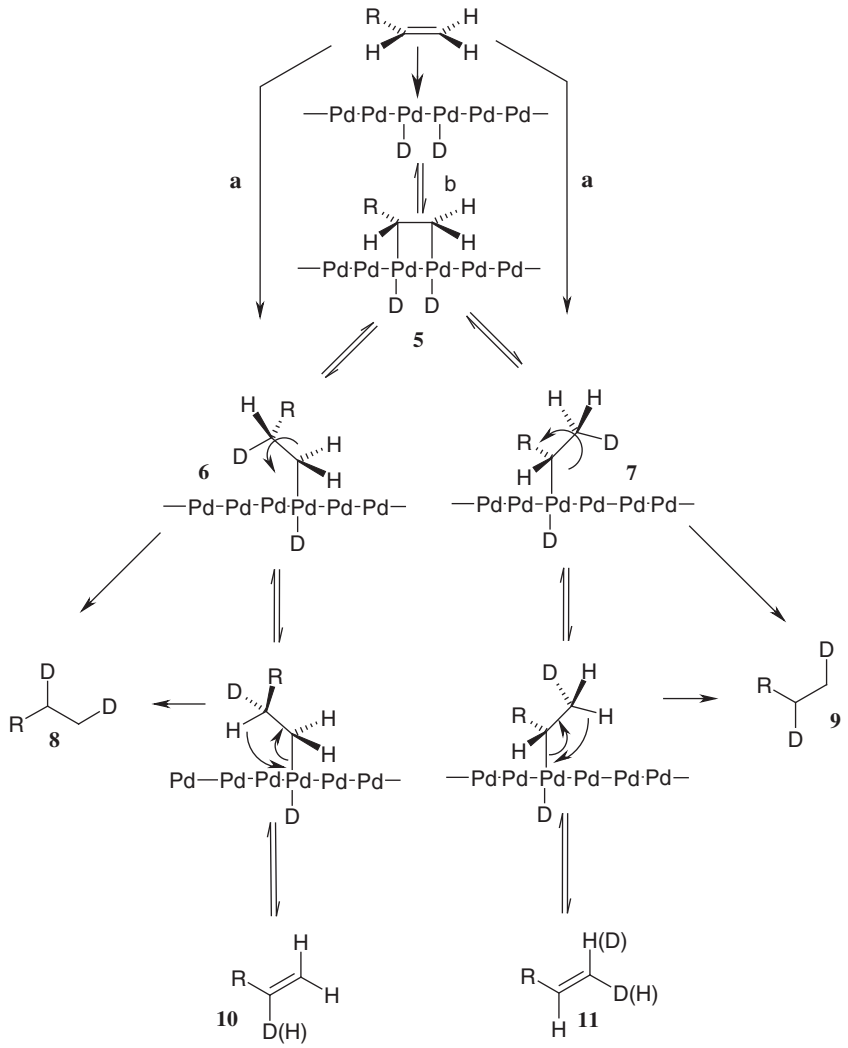


Figure 3. Simplified mechanism for terminal alkene exchange/hydrogenation.

Interestingly, the amphipolar hydropalladation hydrogenation mechanism suggested by Spencer[13a,b] (Route **a**, Figure 3) is also consistent with the preference for terminal over in-chain exchange in the case of mono-substituted terminal alkenes.

Platinum catalysts, however, proved significantly different from all other catalysts in this screen in that they demonstrated unequal degrees of exchange for the terminal *cis* and *trans* protons (the *trans* labelling was significantly preferred) whilst retaining the preference for terminal over in-chain exchange (secondary metal bonding still preferred?).

Moreover, very significant deviations in the preference for terminal vs in-chain labelling were also seen for some mononuclear homogeneous catalysts (little selectivity for secondary metal alkyl species over primary alkyl species or in one case a reversal of selectivity). In these systems di-σ-bonding is very unlikely. This difference could indicate that the di-σ-bonded intermediate **5** has a role in the heterogeneous cases.

The above studies were facilitated by the use and evaluation of a novel ^{3}H-NMR cryoprobe[1a] which enabled hydrogenation and exchange studies to be carried out on a millimolar scale with low specific activity deuterium tritide.[1i,9]

Acknowledgements: In addition to the constant support of John Jones, the following persons/organisations are gratefully acknowledged; EPSRC, ATHENA Johnson Matthey, Stathis Alexakis. David Hesk, James Bloxsidge, Traci Smith, Ed Spink, Stuart Thompson, David Wilkinson, Michael Hickey and the radiochemistry group at AstraZeneca Charnwood.

References

[1] (a) E. Alexakis, J. P. Bloxsidge, J. R. Jones, W. J. S. Lockley, *J Label Compd Radiopharm*, **2006**, *49*, 406–407 and *Synth Appl Isotop Lab Comp, Proc 9th Int Symp, Edinburgh, 2006*, Eds C. L. Willis, W. J. S. Lockley, *J Label Compd Radiopharm*, **2007**, *50*, 300–302; (b) W. J. S. Lockley, D. J. Wilkinson, J. R. Jones, *J Label Compd Radiopharm*, **2006**, *49*, 401–402; (c) M. J. Hickey, J. R. Jones, L. P. Kingston, W. J. S. Lockley, A. N. Mather, D. J. Wilkinson, *Synthesis and Applications of Isotopically Labelled Compounds 2003, Proc. 8th Int. Symp*, (Eds; D. C. Dean, C. N. Filer, K. E. McCarthy), John Wiley, **2004**, *8*, 243–244; (d) J. R. Jones, W. J. S. Lockley, S. Y. Lu and S. P. Thompson. *Tetrahedron Letters,* **2001**, *42*, 331–332; (e) A. S. Culf, H. Morimoto, P. G. Williams, W. J. S. Lockley, W. U. Primrose, J. R. Jones, *J. Lab. Comp. Radiopharm.*, **1996**, *38*, 373–384; (f) W. J. S. Lockley, K. Sfyrakis, B. J. Howlin, J. R. Jones, D. J. Wilkinson, *Synth Appl Isotop Lab Comp, Proc 9th Int Symp, Edinburgh, 2006*, Eds C. L. Willis, W. J. S. Lockley, *J Label Compd Radiopharm*, **2007**, *50*, 532–534; (g) W. J. S. Lockley, R. Sherhod, *J Label Compd Radiopharm*, **2008**, *51*, 258; (h) Analysis of Labelled Compounds by Mass Spectroscopy in the Presence of Heavy Isotopes at Natural Abundance: The NAIC Program, K. N. D. Barakzai, B. J. Howlin, W. J. S. Lockley, K. Sfyrakis, *J Label Compd Radiopharm*, submitted June 2009; (i) E. Alexakis, Ph. D. Thesis, University of Surrey (2005); (j) A. Culf, Ph. D. Thesis, University of Surrey 1994.

[2] (a) W. J. S. Lockley, *Tetrahedron Letters*, **1982**, *23*, 3819–3822; (b) *Synth Appl Isotop Labelled Compds Proc 1st Int Symp, Kansas City*, (Eds; W. P. Duncan, A. B. Susan), Elsevier **1983**, 427–428; (c) W. J. S Lockley, *J Chem Res (S)*, **1985**, 178–179, *J Chem Res* (*M*), **1985**; 1976–1986; (d) W. J. S. Lockley, D. J. Wilkinson, *J Label Compd Radiopharm* **1984**, *21*, 363–373; (e) D. J. Wilkinson, W. J. .S Lockley, *J Label Compd Radiopharm*, **1985**, *22*, 883–892; (f) D. Hesk, J. R. Jones, W. J. S. Lockley, *J Label Compd Radiopharm*, **1990**, *28*, 1427–1436; (g) D. Hesk, J. R. Jones, K. Nolan, W. J. S. Lockley, *Synth Appl Isotop Label Compds Proc 4th Int Symp 1991*, (Eds) E. Buncel, G. W. Kabalka, Elsevier, Amsterdam, **1992**; *4*, 600–603; (h) D. Hesk, J. R. Jones, W. J. S. Lockley, *J Pharm Sci*, **1991**, *80*, 887–890.

[3] (a) L. P. Kingston, W. J. S. Lockley, A. N. Mather, S. P. Thompson, D. J. Wilkinson. *Synthesis and Applications of Isotopically Labelled Compounds 2000, Proc. 7th Int. Symp* (Eds; U. Pleiss, R. Voges), John Wiley **2001**; 139–142; (b) B. M. McAuley, M. J. Hickey, L. P. Kingston, J. R. Jones, W. J. S. Lockley, A. N. Mather, E. Spink, S. P. Thompson, D. J. Wilkinson. *J Label Compd Radiopharm*, **2003**, *46*, 1191–1204; (c) M. J. Hickey, J. R. Jones, L.P. Kingston, W. J. S. Lockley, A. N. Mather, B. M. McAuley, D. J. Wilkinson. *Tetrahedron Lett*, **2003**, *44*, 3959–3961; *J Label Compd Radiopharm*, **2005**, *48*, 75–84; (d) M. J. Hickey, J. R. Jones, L. P. Kingston, W. J. S. Lockley, A. N. Mather, B. M. McAuley, D. J. Wilkinson, *Synthesis and Applications of Isotopically Labelled Compounds 2003, Proc. 8th Int. Symp* (Eds; by D.C. Dean, C. N. Filer, K. E. McCarthy), John Wiley **2004**: 245–246.

[4] (a) E. Alexakis, J. R. Jones, W. J. S. Lockley, *Tetrahedron Lett,* **2006**, *47*, 5025–5028; (b) See the contribution to this symposium by S. C. Schou; (c) S. C. Schou, *J Label Compd Radiopharm*, **2009**, *52*, 376–381.

[5] (a) E. Alexakis, M. J. Hickey, J. R. Jones, L. P. Kingston, W. J. S. Lockley, A. N. Mather, T. Smith, D. J. Wilkinson, *Tetrahedron Lett*, **2005**, *46*, 4291–4293, (b) See e.g. D. Hesk, D. Koharski, S Saluja, P. McNamara, C. Magatti, D. Cesarz, S. Hendershot, J. R. Jones, *Synth. Appl. Isotop. Lab. Comp. Proc. Int. Symp*; (Eds; J. R. Heys, D.G. Melillo, D.G. John Wiley & Sons Ltd), **1998**, *6*, 439–443.

[6] (a) M. J. Hickey, L. P. Kingston, J. R. Jones, W. J. S. Lockley, A. N. Mather, D. J. Wilkinson. *Tetrahedron Lett*, **2004**, *45*, 8621–8623; (b) M. J. Hickey, L. P. Kingston, W. J. S. Lockley, A. N. Mather, D. J. Wilkinson. *J Label Compd Radiopharm*, **2006**, *49*, 388–389; (b) Chemical Week; 9/13/2000; *162* (34), 56–55; http://www.jmcatalysts.com/pharma/index.asp?id = 4 [25 April 2009].

[7] J. R. Jones, A. P. Sharratt, S. D. Jackson, R. W. Griffiths, L. F. Gladden, F. J. Robertson, G. Webb, *J Radioanal Nucl Chem Lett*, **1992**; *166*, 69–74.

[8] http://gow.epsrc.ac.uk/ViewGrant.aspx?GrantRef = GR/R50899/[01 April 09]

[9] E. Alexakis, J. R. Jones W. J. S. Lockley. in *Synth Appl Isotop Label Compds, Proc 9th Int Symp, Edinburgh, 2006*, (Eds; C. L. Willis, W. J. S. Lockley), *J Label Compd Radiopharm*, **2007**, *50*, 507–508.

[10] A. Ozaki, *Isotopic studies of heterogeneous catalysts*, Kodansha (Tokyo) & Academic Press (N York) **1977**, *93*, and references therein.

[11] F. Zaera, *Catalysis Lett*, **2003**, *91*, 1–10.

[12] B. Brandt, J.–H. Fischer, W. Ludwig, J. Libuda, F. Zaera, S. Schauermann, H-J. Freund, *J Phys Chem C,* **2008**, *112*, 11408–11420; A.M. Doyle, Sh.K. Shaikhutdinov, H-J. Freund, *J Catal.* **2004**, *223*, 444–453.

[13] (a) J. Yu, P-S. Whitney, J. B. Spencer, *J Mol Cat A*, **1999**, *146*, 199–210; (b) J. Yu, J. B. Spencer, *Tetrahedron*, **1998**, *54*, 15821–15832 and references therein.

THIRTY YEARS OF TRITIUM LABELING EXPERIENCE AND BEYOND: A LEGACY PASSED DOWN FROM PROFESSOR JOHN R. JONES

YUI S. TANG[#] AND JONATHAN HO

Merck Research Laboratories, Rahway, NJ 07065

[#]Present address: Perkin Elmer LAS, 549 Albany Street, Boston, MA 02118, USA

Abstract: Tritium is the most versatile radio-nuclide used in pharmaceutical researches, and Professor John R. Jones was an undisputable leader in this area. For over forty years, from the time Professor Jones started his interest in the field of isotope chemistry as a post-graduated student and continued to influence the tritium labeling researches since his retirement and untimely

Journal of Labelled Compounds and Radiopharmaceuticals

death, he dedicated his time to the isotope world. Professor Jones not only taught physical organic chemistry and tritium labeling techniques in the University of Surrey with emphasis on understanding of isotope effects and mechanisms, but lead a very active group to carry out cutting edge researches of tritiation labeling methods. The authors, one of his many students and another one of his scientific admirers respectively, detail some personal encounters of his research activities and how his work and ideas shaped modern tritium labeling methodology.

Keywords: Tritium Labeling; Catalysis; Tritium NMR

Results and Discussion: In 1979 when John recruited the author (YST) under his wing as a CASE (Industrial) doctorate student, the first task was to solve an unexplained unevenly tritium labeling across the carbon double bond of dehydro-alprenolol. John, who collaborated with the late Professor John Elvidge (then the Head of Chemistry Department at University of Surrey), and late Professor Antony Evans (Amersham International Plc), was a pioneer and expert in Tritium NMR spectroscopy and was greatly puzzled by the relative 1:3 ratio of the labeled methylene and methyl groups (Figure 1). We speculated the heterogeneous catalytic activities on the surface during the association of tritium gas resulted in the uneven incorporation of tritium. Our approach was to study the hydrogenation step-wise using less than one equivalent of tritium gas to reduce the double bond(deficiency of tritium gas), isolate the reductive intermediates, and examined the Tritium NMR spectra. During the study, all the terminal vinylic protons underwent even triton-proton exchange prior to the reduction. We named this intermediate step 'Vinylic Exchange Mechanism' (Figure 2). From the precise TNMR interpretation of the relative tritium distribution, we concluded that the fully reduced C-C double bond produced the relative 1:3 tritium ratios in favor of the terminal methyl group. In addition, double bond migration-exchange contributed to scrabble of tritium atoms at the methine position. Theoretical calculations of labeled species 70:25:5, corresponds to the total distribution of tritium of the dihydroalprenolol.

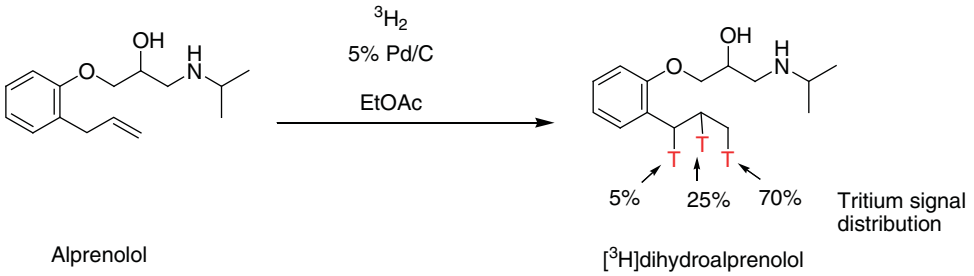


Figure 1. Heterogeneous catalytic reduction of alprenolol showing distribution of tritium[1].

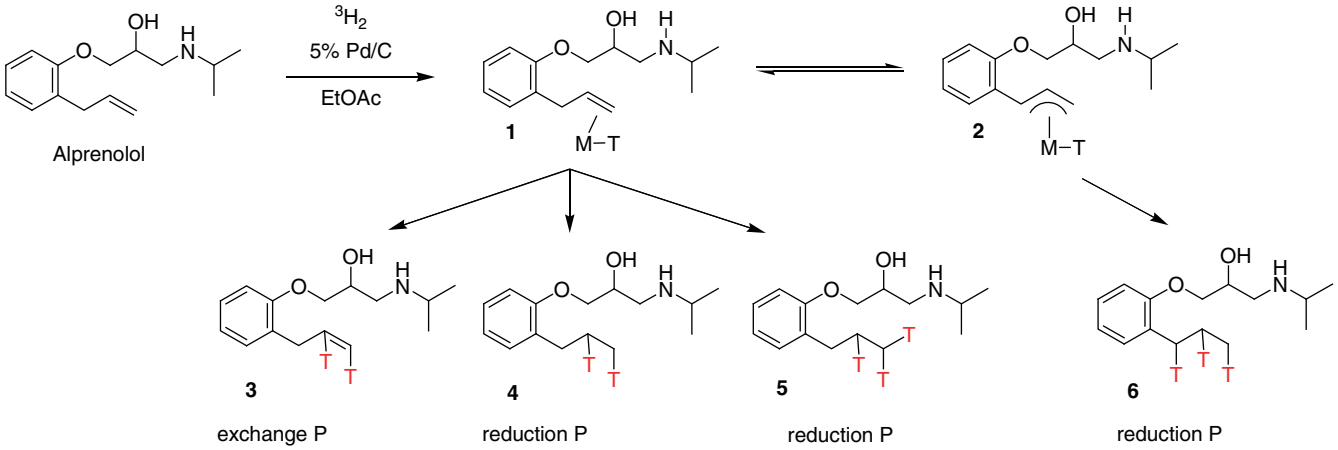


Figure 2. Reduction of unsaturated bond via vinylic exchange and double-bond migration mechanism[2].

We made practical use of this newly discovered mechanism and we prepared some vinylic exchanged tritium labeled biological e.g. [^{3}H]Kainic acid and [^{3}H]Isosafrole[3]. The isotope world quickly adopted this method to prepare some of the vinylic exchange label compounds as exemplified by the elegant synthesis of tritiated Cyclosporin-A by R. Voges's group at Sandoz [4].

Catalytic dehalogenation reaction has been a useful tool for the incorporation of tritium into organic chemicals, but sometimes produces very puzzling results as the unexpected lower than theoretical specific activity and some tritium eventually ends up different positions then the halogenated position. Debromination of dibromfolic acid is an extensively studied process [5]. Under

John's guidance, we investigated different catalysts and solvent conditions but we failed to find a rationale for the dehalogenation reaction. We also were not able to do selective dehalogenation in the presence of other reducible groups. These problems were not solved until twenty years later using sodium borotritide and transition metal catalysts (Figure 5). Nevertheless, experience and understanding for such process were acquired for the later success.

John was known as a competent radiochemist but there are not a lot of references on how innovative and technical skilled researcher he was. In the summer of 1981, Professor M. Long from University of New South Wales, Australia and Phil Williams spent three months on a collaborate research project in our laboratory. We designed a simple home-made GC radio-detector to analyze reaction mixtures [6]. This device allowed us a quick and reliable method to scan catalytic effects of dehalogenation and double-bond and triple bond reduction reactions in order to optimize our reactions.

John had much influence on the isotope world with collaborative research activities with Professor Burcell's (University of Kingston, Ontario) group. They served as long term co-editor of the Journal of Labelled Compounds and Radiopharmaceuticals and also co-edited many books on radioisotope techniques and isotope labelings. John worked closely with Professor A. Jerry Kresge (University of Toronto, Ontario, Canada) and sent post-doctor students to Canada. Jerry and John had mutual respect to each other and the two groups published many papers on kinetic isotope researches on [7a–c]

Pka and Keto-enol Equilibrium Constant of Acetone in Aqueous Solution
Tritium Isotope Effects on C-13 NMR Shifts
The ionization of Terminal Acetylenes: Pseudo-Acid Behavior
The Acidity of the Terminal Acetylene Proton in the Studies of De-tritiation
Relationship between the Acidic Proton in Some Potent General Anesthetics

One of the most important projects was the kinetic isotope effect on some general anesthetic agents. The relationship between the acidity of the lone proton in some of the most potent general anesthetic agents could be a virtual tool in the investigation of more controllable new potent general anesthetics. Also understanding of physical organic solution kinetics could be extrapolated using the hydrogen isotope effects [8].

John's influence on the technique of tritium reactions spread to the US when some of the top-notched tritium research programs were completed by some of his students who went to the National Tritium Labeling Facilities, University of California, Berkeley. His students included Y. S. Tang, M. Saljoughian, D. Java and P .Williams who made major contributions in improving tritiation reactions[9]. e.g.

Tritiation technology: Catalytic hydrogenation and dehalogenation
Recovery tritium waste: catalytic converter from tritium gas to tritiated water
Development of Uranium tritium storage technology
Microwave and solid phase tritiation of proteins and peptides
Development of super active tritiated methyl iodide and tritides
Tritium NMR spectroscopy
Tritium detection systems for GC and HPLC

From 1990–2008, John's influence on the tritiation technologies blossomed and many new methods and novel tritium reagents have been prepared successfully. Figures 3–7 highlight some of the useful methods that have shaped tritium labeling technology and John's isotope influence.

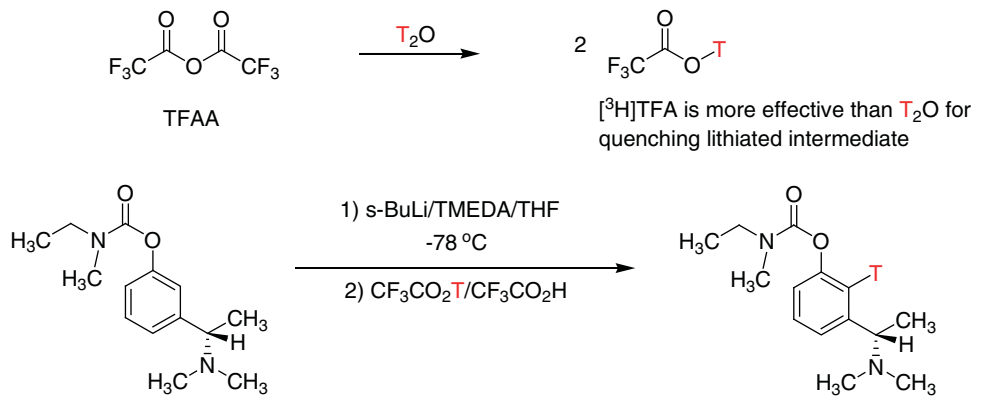


Figure 3. Tritium labeling of acetylcholine esterase inhibitor via ortho-metalation and tritiation with TFAA and T_2O[10].

Journal of Labelled Compounds and Radiopharmaceuticals

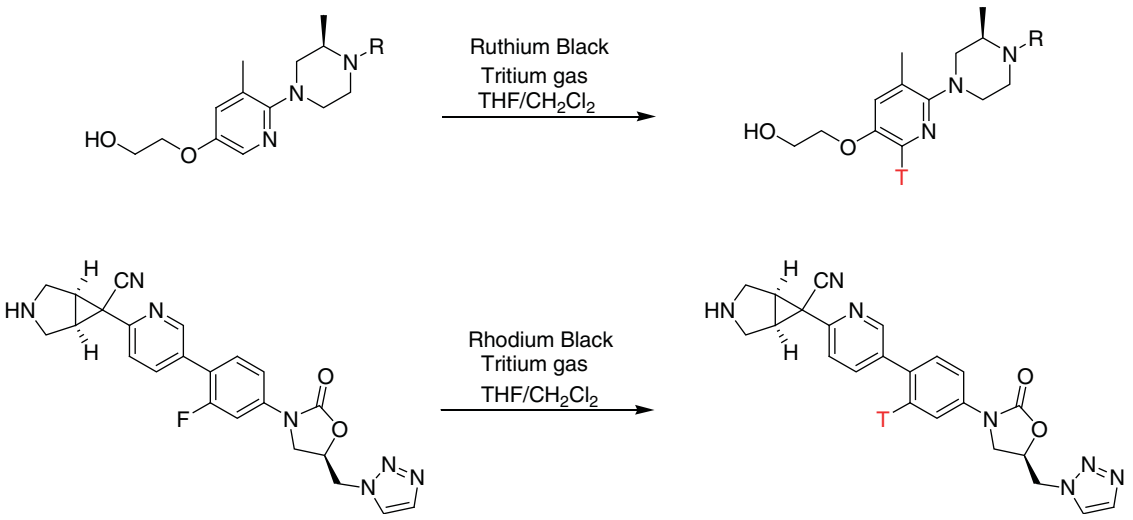


Figure 4. Labeling pyridines using a heterogeneous catalytic exchange method developed by Professor Jones and colleagues at University of Surrey[11].

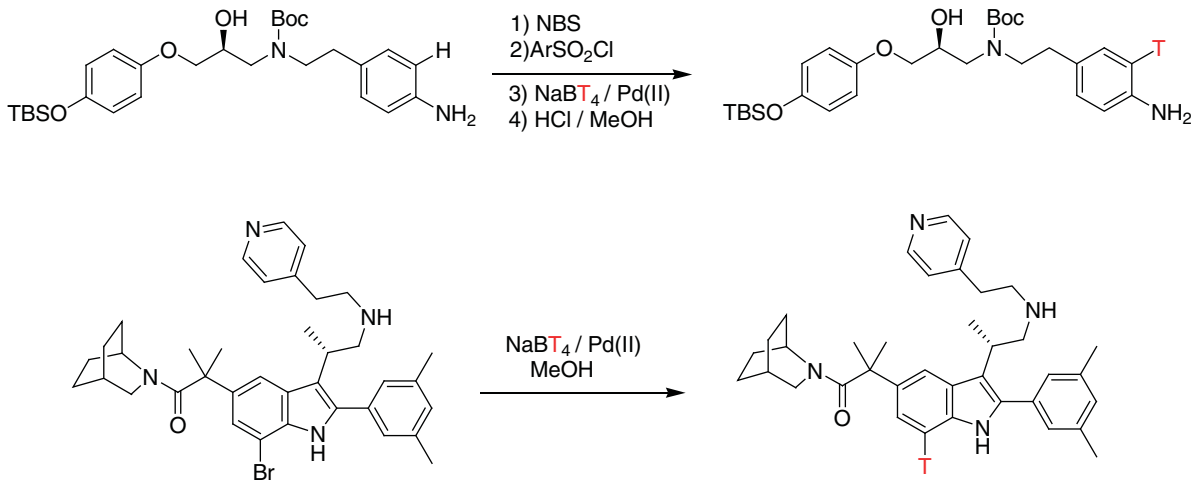


Figure 5. Regio-specific tritio-dehalogenation labeling method using $NaBT_4$/Pd(II) catalyst[12].

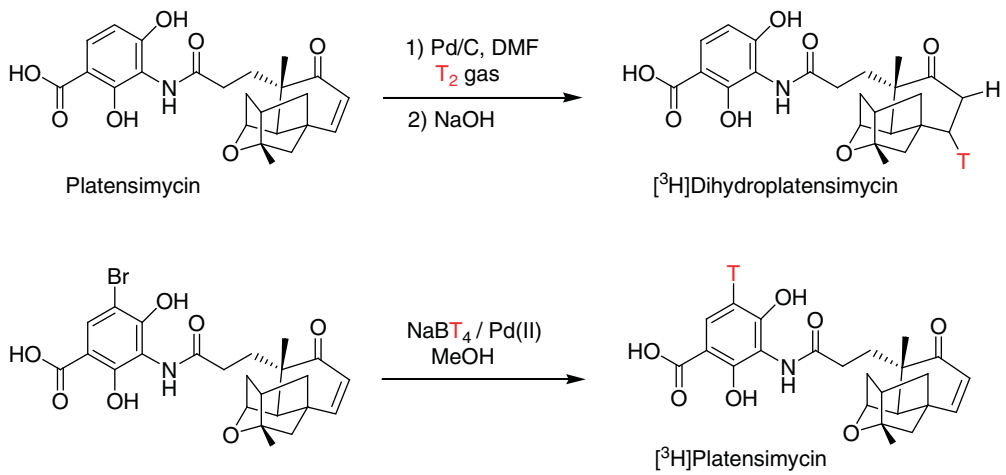


Figure 6. Regio-specific and selective tritio-dehalogenation reaction for Platensimycin[13].

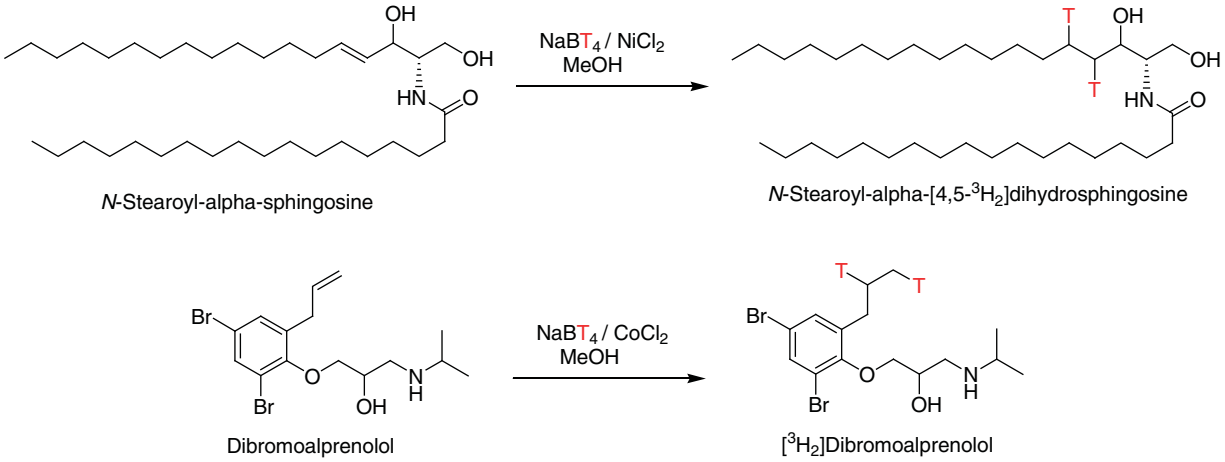


Figure 7. Regio-specific tritiation of a carbon-carbon double bond using $NaBT_4$/Ni (II) and CO(II) transition metal catalysts[14].

References

[1] J. A. Elvidge, J. R. Jones, R. M. Lenk, Y. S. Tang, E. A. Evens, G. L. Guilford and D. C. Warrell, *J. Chem. Research* (S) **1982**, 82–83.
[2] J. A. Elvidge, J. R. Jones, Y. S. Tang, E. A. Evens, G. L. Guilford and D. C. Warrell, Proceedings of the 3rd IIS meeting, Kanas City, **1983**, 327–330.
[3] Yui S Tang, Ph.D. Thesis, University of Surrey, UK, **1982.**
[4] R. Voges, Private communication.
[5] J. A. Elvidge, J. R. Jones, R. M. Lenk, E. A. Evens, G. L. Guilford and D. C. Warrell, *J. Label Compd & Radiopharm*, **1979**, 16(5), 697–701.
[6] J. R. Jones, M. Long, P. G.Willaims and Y. S. Tang, *J. of chromatography*, **1984**, *287*, 381–384.
[7] (a) A. J. Kresge, Y. Chiang, Y. S. Tang and *J. Wirz*, *JACS*, **1984**, *106*, 460–464; (b) A. J. Kresge, Y. S. Tang and M. F. Powell, JACS, **1986**, 108–120; (c) A. J. Kresge, Y. S. Tang, JACS, **1987**, *109*, 7108–7112.
[8] A. J. Kresge, Y. Chiang and Y. S. Tang, JACS, **1989**, *111*, 4918–4921.
[9] P. G. Williams, Y. S. Tang and H. Morimoto Chemistry in Australia, March, **1988**, 75–77.
[10] G. Ciszewska, Y. S. Tang and U. Sunay, J. Label Compd & Radiopharm, **1997**, *39*(8), 652–658.
[11] J. R. Jones, W. J. Lockley and D. J. Wilkinson, *J. Label Compd & Radiopharm*, Synthesis and Application of Isotopically labeled compounds, **2005**, *48*, 523–524.
[12] A. E. Weber, Y. S. Tang et al, *Bio-organic and Medicinal Chemistry Letters*, **1998**, *8*, 2111–2118.
[13] Jun Wang, Y. S. Tang et al, *Nature*, **2006**, *441*, 358–363.
[14] Unpublished results, Merck and Co.

FLUORINE-18 CHEMISTRY IN MICRO-REACTORS

SHUIYU LU, JOONG-HYUN CHUN AND VICTOR W. PIKE

Molecular Imaging Branch, National Institute of Mental Health, National Institutes of Health, 10 Center Drive, Room B3 C346, Bethesda, MD 20892-1003, USA

Abstract: Recent applications of micro-reactor (microfluidics) technology to radiofluorination chemistry within our laboratory are presented, based on use of either a simple T-shaped glass micro-reactor or a more advanced microfluidics instrument. The topics include reaction optimization and radioligand production, in particular the study of the radiofluorination of diaryliodonium salts, [^{18}F]fluoride ion exchange with xenon difluoride, esterification with [^{18}F]2-fluoroethyl tosylate, and the syntheses of [^{18}F]fallypride, [^{18}F]FBR and [^{18}F]SL702 from [^{18}F]fluoride ion.

Keywords: micro-reactor; microfluidics; fluorine-18; fluorination; PET

Introduction: It is increasingly recognized that micro-reactor (or microfluidics) technology can provide considerable advantages in radiochemistry with short-lived positron-emitting fluorine-18 ($t_{1/2}$ = 109.7 min).[1–5] Possible benefits to be derived from this technology include: (1) the use of smaller amounts of materials, especially non-radioactive precursor, which may be precious or difficult to obtain; (2) easier and more efficient radioactive product purification; (3) reduced radiation exposure to radiochemists by allowing more efficient radiosynthesis with less radioactivity; and (4) potential for scale-up and a high degree of automation.

Journal of Labelled Compounds and Radiopharmaceuticals

We have used micro-reactor technology to optimize radiofluorination procedures and to produce radioligands reliably and reproducibly for molecular imaging in rodents with positron emission tomography (PET).[6–15] Other groups are also exploring microfluidics in PET chemistry.[16–20] This presentation tracks the growth of our research in this area covering the early development of a simple glass T-shaped micro-reactor to recent progress in several areas of fluorine-18 chemistry in a more advanced microfluidics instrument. Our examples show-case some of the various benefits of using microfluidics in fluorine-18 chemistry.

Results and Discussion: *[^{18}F]3-(3-Pyridinyl)propionic-2′-fluoroethyl ester by esterification*

As a proof-of-principle study, our first examples of radiosyntheses with a simple hydrodynamically-driven glass micro-reactor were those of ^{18}F-labeled esters (**Figure 1**).[6] 2-[^{18}F]Fluoroethyl esters are sometimes proposed as PET radiotracers.[21] Such esters may be prepared by reactions of carboxylic acid salts with [^{18}F]2-fluoroethyl tosylate (**4**). The synthesis of the model 2-fluoroethyl ester **3** in moderate yield from the reaction of **1** with 2-fluoroethyl tosylate (**2**) required warming the micro-reactor to 80°C. Yields were dependent on reactant concentration and flow rate. At lower flow rate, a higher concentration of reactant **1** gave more ester **3**. The reaction could be performed with as low as 0.75 μg (5 nmol) of **1** in 10 μL of solution. Reactions of **1** with the labeling agent **4** at 80°C, at infusion rates of 1 μL/min, gave the corresponding ^{18}F-labeled ester **5** in 10% decay-corrected radiochemical yield (RCY). This device had however limited scope for control of temperature and reaction stoichiometry. Radiosyntheses were necessarily carried out at <2 mCi level because the procedure was not highly automated.

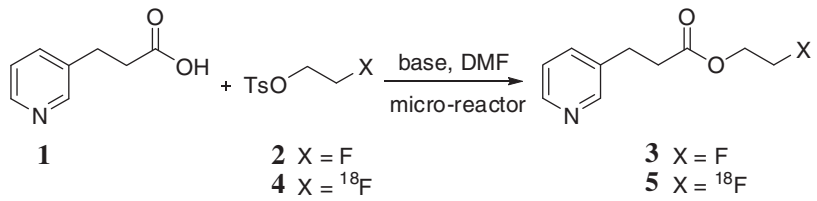


Figure 1. Labeling of **5** with [^{18}F]2-fluoroethyl tosylate in a simple micro-reactor.

[^{18}F]Fallypride by aliphatic nucleophilic substitution

A commercial coiled-tube micro-reactor (NanoTek; Advion) subsequently became a convenient platform for the study of ^{18}F-labeling with microfluidics in our laboratory. Radiosynthesis of the brain dopamine D_2 receptor radioligand, [^{18}F]fallypride (**7**)[22] from the tosylate-precursor **6** was rapidly optimized in this apparatus, with respect to the effects of precursor amount, reaction temperature, flow rate and [^{18}F]fluoride ion to precursor concentration ratio (**Figure 2**).[7] Each radiosynthesis used low amounts (2040 μg; 3977 nmol) of **6** and [^{18}F]fluoride ion/K 2.2.2 (0.52.5 mCi). RCYs of **7** (up to 88%) were reproducible. The low amounts of material used in each radiosynthesis allowed crude **7** to be purified rapidly on an analytical-size reversed phase HPLC column, preceding formulation for intravenous injection. Scale-up of the reaction was achieved by continuously infusing precursor and [^{18}F]fluoride ion solutions into the reactor to obtain **7** in much greater radioactivity (>10 mCi). In this instrument, **7** was conveniently synthesized in small doses (0.31.5 mCi) for micro-PET studies in rodents.

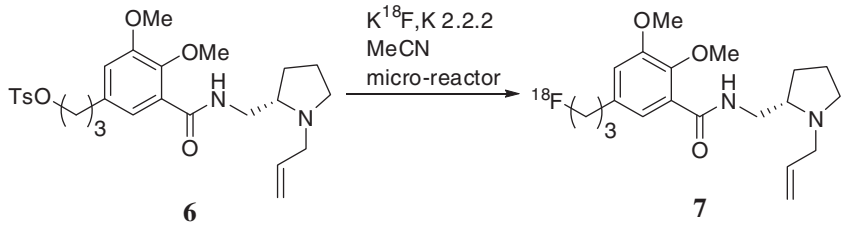
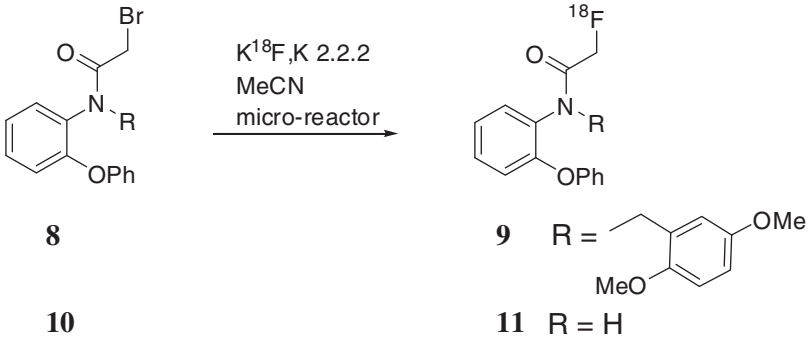


Figure 2. Synthesis of **7** in a micro-reactor.

[^{18}F]FBR by aliphatic nucleophilic substitution

[^{18}F]FBR (**9**) is an effective TSPO radioligand[8] which is now being used to study inflammatory conditions in human subjects.[23,24] We have shown that **9** can be produced in high RCY in the Advion microfluidic apparaus (**Figure 3**). Below 90°C the RCY of **9** was lower in slightly wet acetonitrile than in anhydrous acetonitrile. However, this difference in RCYs disappeared when the reactor temperature was raised above 110°C. RCYs reached 85% at 110°C. Further temperature increase gave no improvement in RCY. The synthesis of the main metabolite (**11**) of **9** was also investigated in the microfluidic apparatus under similar conditions. The RCY was substantially lower than for **9** over the temperature range 30-150°C. **9** and **11** were each prepared with this microfluidic apparatus in sufficient amounts for intravenous injection into rodents.



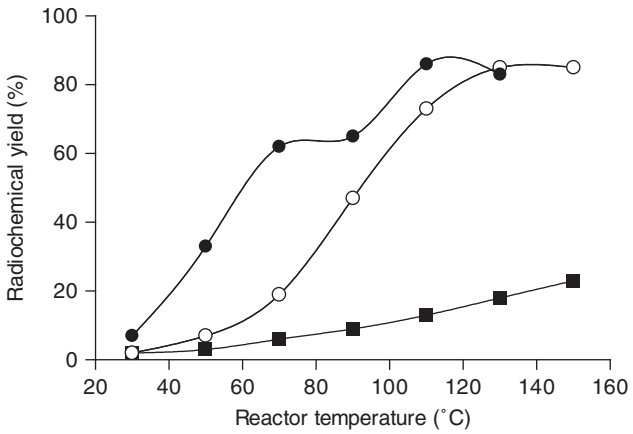


Figure 3. Synthesis of **9** and **11** in a micro-reactor and temperature dependence of RCYs of **9** in anhydrous acetonitrile (●) or acetonitrile with 0.3% v/v water (o), and of **11** in anhydrous acetonitrile (■).

[^{18}F]SL702 synthesis by flow or stopped-flow mode

[^{18}F]SL702 (**14**), a new potential agonist radioligand for brain 5-HT$_{1A}$ receptors, was successfully prepared in the Advion micro-reactor from a nitro-precursor (**12**) in a moderate RCY, similar to that obtained in a conventional microwave procedure (Route A, **Figure 4**).

Direct ^{18}F for ^{19}F exchange could be a useful route for labeling with fluorine-18 where the target radiotracer is not required to have high specific radioactivity. Although, high specific activity would be required for applications of [^{18}F]SL702, we wished to test the feasibility of performing ^{18}F for ^{19}F exchange for this type of structure. We first investigated the exchange reaction using a low concentration of **13** (Route B, **Figure 4**). When the flow rate was 5 μL/min, the RCY was about 4%. A 'stopped flow' method was introduced to allow longer reaction time in the micro-reactor. For 10 min reaction time the RCY increased to 11%. Further optimization for this and other targets is under investigation.

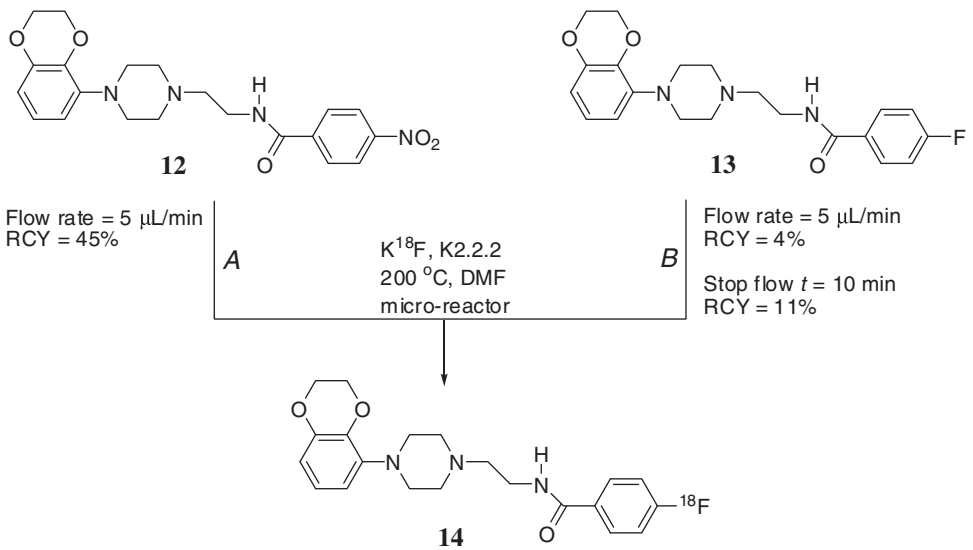


Figure 4. Synthesis of **14** in a micro-reactor by aromatic nucleophilic substitution in a nitro-precursor (Route A) or from ^{18}F for ^{19}F exchange (Route B).

Journal of Labelled Compounds and Radiopharmaceuticals

Syntheses of [^{18}F]fluoroarenes from diaryliodonium salts

Reactions of diaryliodonium salts with [^{18}F]fluoride ion are increasingly useful for the preparation of [^{18}F]fluoroarenes as radiotracers from NCA [^{18}F]fluoride ion.[25,26] The Advion micro-reactor apparatus has enabled us to study the RCYs, product selectivity, kinetics and energetics of these reactions in detail (**Figure 5**).[10–14] This platform was very convenient for running multiple reactions rapidly under well-controlled conditions of reactant concentrations, reaction times and reaction temperatures. High RCYs of a vast array of NCA [^{18}F]fluoroarenes were obtained in short reaction times (<7 min).

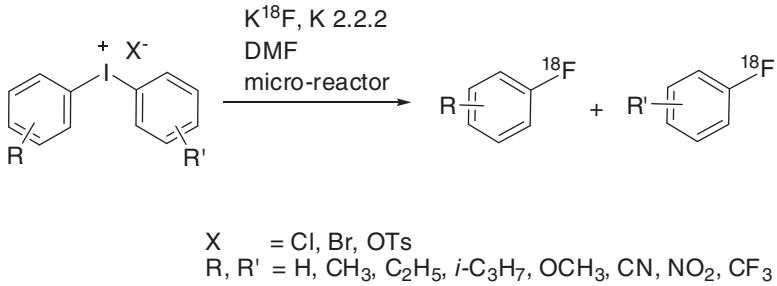


Figure 5. Reactions of diaryliodonium salts with [^{18}F]fluoride ion to produce [^{18}F]fluoroarenes in a micro-reactor.

[^{18}F]XeF_2 by direct exchange

^{18}F-Labeled xenon difluoride ([^{18}F]XeF_2) is a potentially useful 'electrophilic' radiofluorination agent. We have previously prepared [^{18}F]xenon difluoride by exchange of xenon difluoride with [^{18}F]fluoride ion (as $^{18}F^-Cs^+$/K 2.2.2) in dichloromethane at room temperature (**Figure 6**).[27] This process is attractive because of the availability of [^{18}F]fluoride ion in high activity and high specific activity from the ^{18}O(p,n)^{18}F reaction on [^{18}O]water. We exploited a microfluidic device to study further the influence of different conditions on this reaction.[15] [^{18}F]Xenon difluoride was obtained in high RCY (50%) by exchange of [^{18}F]fluoride ion with xenon difluoride in either dichloromethane or acetonitrile. In acetonitrile, the reaction may be performed in the presence of Cs^+, with or without K 2.2.2, or with K^+-K 2.2.2 at elevated temperature. The use of a micro-reactor allowed close monitoring of the progress of the exchange reaction with time and temperature. The procedure has potential to produce [^{18}F]xenon difluoride consistently and rapidly, and at a usefully high specific radioactivity.

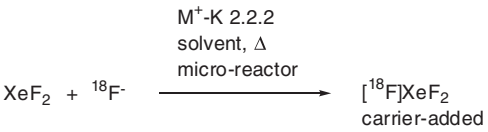


Figure 6. Preparation of [^{18}F]xenon difluoride by exchange with [^{18}F]fluoride ion in a micro-reactor.

Conclusions: Research in our laboratory and elsewhere has clearly demonstrated that micro-reactor or microfluidics technology is well suited to PET radiotracer development and production. Our results exemplify some of the potential advantages of this methodology for radiotracer development and synthesis. This technology should be increasingly amenable to greater sophistication to encompass entire radiosyntheses in a versatile high throughput manner and should play a more significant role in advancing PET radiochemistry.

Acknowledgements: This research was supported by the Intramural Research Program (Project No Z01-MH-002793) of the NIH, NIMH. SYL and VWP dedicate this presentation to the memory of Professor John R. Jones for invaluable mentorship and friendship in former times.

References

[1] S. Y. Lu, V. W. Pike, in *PET Chemistry– The Driving Force in Molecular Imaging*, (Eds: Schubiger PA, Lehmann L, Friebe M), Springer-Verlag, Heiderberg, **2007**, pp. 271–287.
[2] G. Lucignani, *Eur. J. Nucl. Med. Mol. Imaging*, **2006**, *33*, 849–851.
[3] L. S. Cai, S. Y. Lu, V. W. Pike, *Eur. J. Org. Chem*., **2008**, *17*, 2853–2873.
[4] P. W. Miller, *J. Chem. Technol. Biotechnol.*, **2009**, *84*, 309–315.
[5] A. Elizarov, *Lab Chip*, **2009**, *9*, 1326–1333.
[6] S. Y. Lu, P. Watts, F. T. Chin, J. Hong, J. L. Musachio, E. Briard, V. W. Pike, *Lab Chip*, **2004**, *4*, 523–525.
[7] S. Y. Lu, A. M. Giamis, V. W. Pike, *Curr. Radiopharm*., **2009**, *2*, 49–55.
[8] E. Briard,S. S. Zoghbi, F. G. Siméon, M. Imaizumi, G. P. Gourley, H. U. Shetty, S. Y. Lu, M. Fujita, R. B. Innis, V. W. Pike, *J. Med. Chem*., **2009**, *52*, 688–699.
[9] S. Y. Lu, J. T. Clements, M. J. Gilde, A. Prak, P. Watts, V. W. Pike, *J. Labelled Compd. Radiopharm*., **2007**, *50*, 597–599.
[10] J. Chun, S. Lu, V. W. Pike, *J. Labelled Compd. Radiopharm*. **2009**, *52* (Suppl. 1): S2.
[11] S. Telu, J. Chun, F. G. Siméon, S. Y. Lu, V. W. Pike, *J. Labelled Compd. Radiopharm*. **2009**, *52* (Suppl. 1): S4.

[12] J. Chun, S. Lu, V. W. Pike, *J. Labelled Compd. Radiopharm*. **2009**, *52* (Suppl. 1): S14.
[13] J. Chun, S. Lu, V. W. Pike, *J. Labelled Compd. Radiopharm*. **2009**, *52* (Suppl. 1): S156.
[14] J. Chun, S. Lu, Y. S. Lee, V. W. Pike, *J. Labelled Compd. Radiopharm*. **2009**, *52* (Suppl. 1): S159.
[15] S. Lu, Y. S. Lee, V. W. Pike, *J. Labelled Compd. Radiopharm*. **2009**, *52* (Suppl. 1): S496.
[16] Abstracts of Presentations, Session 2: Microfluidics and nanotechnology, 18th International Symposium on Radiopharmaceutical Sciences, *J. Labelled Compd. Radiopharm.*, **2009**, *52* (Suppl. 1): S8–S14.
[17] Abstracts of Presentations, Novel technology: nanotechnology, microreactors and microfluidics posters, *J. Labelled Compd. Radiopharm.*, **2009**, *52* (Suppl. 1): S496–S508.
[18] C. C. Lee, G. D. Sui, A. Elizarov, C. Y. J. Shu, Y. S. Shin, A. N. Dooley, J. Huang, A. Daridon, P. Wyatt, D. Stout, H. C. Kolb, O. N. Witte, N. Satyamurthy, J. R. Heath, M. E. Phelps, S. R. Quake, H. R. Tseng, *Science* **2005**, *310*, 1793–1796.
[19] J. M. Gillies, C. Prenant, G. N. Chimon, G. J. Smethurst, W. Perrie, I. Hamblett, B. Dekker, J. Zweit, *Appl. Radiat. Isot,* **2006**, *64*, 325–332.
[20] C. J. Steel, A. T. O'Brien, S. K. Luthra, F. Brady, *J. Labelled Compd. Radiopharm.*, **2007**, *50*, 308–311.
[21] S. Y. Lu, F. T. Chin, J. A. McCarron, V. W. Pike, *J. Labelled Compd. Radiopharm*. **2004**, *47*, 289–297.
[22] J. Mukherjee, Z. Y. Yang, T. Brown, R. Lew, M. Wernick, X. H. Ouyang, N. Yasillo, C. T. Chen, R. Mintzer, M. Cooper, *Nucl. Med. Biol*., **1999**, *26*, 519–527.
[23] M. Imaizumo, E. Briard, S. S. Zoghbi, J. Hong, J. L. Musachio, V. W. Pike, R. B. Innis, M. Fujita, *Synpgapse*, **2007**, *61*, 595–605.
[24] Y. Fujimura, S. S. Zoghbi, F. G. Simèon, A. Taku, V. W. Pike, R. B. Innis, M. Fujita, *J. Nucl. Med.*, **2009**, *50*, 1047–1053.
[25] V. W. Pike, F. I. Aigbirhio, *J. Chem. Soc., Chem. Commun*., **1995**, 2215–2216.
[26] A. Shah, V. W. Pike, D. A. Widdowson, *J. Chem. Soc., Perkin Trans I*, **1998**, 2043–2046.
[27] M. Constantinou, F. I. Aigbirhio, R. G. Smith, C. A. Ramsden, V. W. Pike, *J. Am. Chem. Soc*., 2001, **123**, 1780–1781.

