JOURNAL OF LABELLED COMPOUNDS AND RADIOPHARMACEUTICALS
J Label Compd Radiopharm 2003; **46**: 489–498.
Published online in Wiley InterScience (www.interscience.wiley.com). DOI: 10.1002/jlcr.692

Research Article

Application of 1-butyl-3-methylimidazolium hexafluorophosphate to Ir(I)-catalyzed hydrogen isotope exchange labelling of substrates poorly soluble in dichloromethane

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Summary

Substrate solubility remains a major limitation in Ir(I)-catalyzed isotopic hydrogen exchange labelling. In the search for an alternative to the solvent dichloromethane, which is critical to the success of the reaction, we examined a series of ionic liquids for their suitability. Commercially available 1-butyl-3-methylimidazolium hexafluorophosphate (abbreviated to [BMI][PF_6]) was found to support efficient deuterium and tritium exchange labelling of *N*-(4-methoxyphenyl)-*N*-methyl benzamide **1** under standard conditions. The solvent dissolves both polar hydroxyl and carboxylic acid substituted acetanilides, providing isotopomers in unprecedentedly high deuterium incorporation as compared to dichloromethane. We report the application of [BMI][PF_6] and its potential for extending the scope of Ir(I)-catalyzed H/T exchange to more polar compounds. Copyright © 2003 John Wiley & Sons, Ltd.

Key Words: ionic liquids; iridium catalyzed isotopic hydrogen exchange labelling; alternative solvent to dichloromethane; labelling of polar compounds

Abbreviations: **[EMI]**: 1-ethyl-3-methylimidazolium; **[BMI]**: 1-butyl-3-methylimidazolium; **[HMI]**: 1-hexyl-3-methylimidazolium; **[OMI]**: 1-octyl-3-methylimidazolium; **[NTf_2]**: bis(trifluoromethanesulfonyl) amide; **[OTf]**: trifluoromethanesulfonate; **[TF]**: trifluoroacetate; **PPF**: (*R*)-1-{(1*S*)-2-[bis(4-methoxy-3,5-dimethylphenyl)phosphino]ferrocenyl}

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Received 11 November 2002
Accepted 28 November 2002

ethyldicyclohexylphosphine; **BARf**: tetrakis(3,5-bis(trifluoromethyl)-phenyl)borate; **dppb**: 1,4-bis(diphenylphosphino)butane; **py**: pyridine.

Introduction

Isotopic labelling of biologically active compounds is an essential tool for drug development and basic research in the pharmaceutical industry. From several strategies available, homogeneous catalyzed hydrogen isotope exchange labelling of complex drug substances is appealing because it is a fast, one step reaction leading to isotopomers by simple replacement of hydrogen for deuterium or tritium in the presence of a catalytic amount of an iridium catalyst. The scope and applications of this methodology has been intensively investigated during the past decade.[1–4] The method relies solely on the use of dichloromethane, which is critical to the success of the reaction,[5] in order to support catalytic exchange through what is considered to be a finely balanced ligating and leaving ability with the iridium metal center. Since a large proportion of compounds within pharmaceutical development are essentially insoluble in dichloromethane, there is no choice but to employ a more time consuming synthetic route. Alternatives have been sought such as dimethylformamide, dioxane and acetonitrile,[†2] but due to strong complexation with the active catalyst species, they prevent isotopic exchange processes from taking place. Those that have worked albeit to a lesser extent, such as acetone,[†] ethyl acetate and chlorobenzene,[3] have failed to address the primary goal of solubility. Since ionic liquids are able to dissolve polar compounds and salts, and allow one to moderate their nucleophilic character (and hence their affinity to the electrophilic metal catalyst centre) by appropriate selection of the counterion, they offer the possibility to circumvent the problems associated with traditional highly polar solvents. The need for an additional solvent was also in part driven by emerging evidence for the formation of a presently unidentified non-readsorbable highly tritiated gas formed from reactions involving dichloromethane and iridium catalysts.[‡] This paper describes a two step approach beginning

[†]Presentation given by Heys JR at the Annual International Isotope Society (UK Group) Symposium, May, 1997.

[‡]Tritium exhaust monitoring equipment has recently revealed the production of significant quantities (GBq) of an unidentified non-re-adsorbable tritiated gas species suspected to be formed from reaction of dichloromethane with the iridium (I) catalyst. Further results from these studies will be published later.

with screening to identify a solvent that mimics the potential of dichloromethane followed by application of the most effective medium to the labelling of substrates insoluble in dichloromethane.

Results and Discussion

H/D exchange of N-(4-methoxyphenyl)-N-methyl benzamide **1** *in ionic liquids*

The following reactions were run for 16 h in the presence of 10 mol% Crabtree catalyst under 1 atmosphere deuterium gas at room temperature. For reasons of convenience experiments were run with deuterium instead of tritium, however both gases were found to behave similarly as described later in this article. Table 1 illustrates the results obtained for *N*-(4-methoxyphenyl)-*N*-methyl benzamide **1** in different ionic liquids of the type illustrated in scheme 1.

A, B and C, Denote the percentage deuterium incorporation into the designated positions of **1**, scheme 1. Reactions were run at least twice and deuterium incorporation was calculated from the decrease of the ^{1}H-NMR signal, and data were found to be within $\pm$ 10% tolerance.

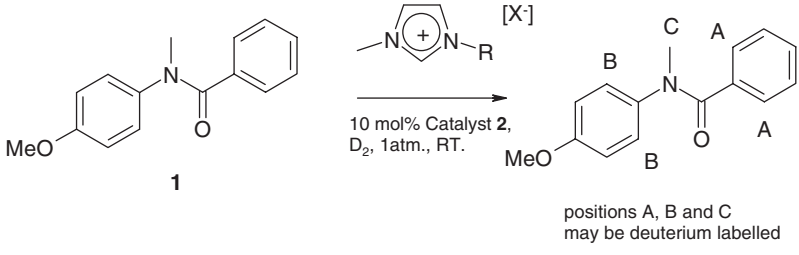
Initial results were very encouraging with iridium-mediated exchange taking place in ionic liquids bearing [PF_6] and [OTf] counterions. The more nucleophilic [TF] and [NTf_2] counterions[6] completely suppressed exchange presumably by blocking the key conversion between the coordinatively unsaturated-saturated catalyst species. Surprisingly, the less nucleophilic [BF_4] analog also failed to mediate catalytic exchange using either commercial or homemade sources. Since purity is reportedly difficult to achieve[7] one explanation may be the presence of trace amounts of highly coordinating chloride ions.

Given the commercial availability of ionic liquids bearing the [PF_6] anion, we focussed the development of the methodology on 1-(R)-3-methylimidazolium hexafluorophosphate ionic liquids and varied the length of substituent, R systematically. Increasing chain length, R, from ethyl, butyl, hexyl to octyl gave a surprising variation in deuterium incorporation. Factors such as viscosity, solubility of substrate, catalyst and deuterium gas, in the ionic liquid influence the net outcome. Both [OMI] and [BMI] ionic liquids gave comparable results to dichloromethane (last entry) however, we chose to employ [BMI][PF_6] in all subsequent studies due to its commercial availability, as well as its lower viscosity and lipophilic character.

Table 1. Deuteration of compound 1 in different ionic liquids at room temperature

Abbreviation	Counterion X	R	A (%)	B (%)	C (%)
[EMI][BF_4][a]	BF_4^-		0	0	0
[EMI][PF_6][c]	PF_6^-	*n*-Ethyl	83[d]	0[d]	12[d]
[EMI][NTf_2][b]	$(CF_3SO_2)_2N^-$		0	0	0
[EMI][TF][b]	$CF_3CO_2^-$		0	0	0
[BMI][BF_4][b]	BF_4^-		0	0	0
[BMI][PF_6][a,b]	PF_6^-		90	19	20
[BMI][NTf_2][b]	$(CF_3SO_2)_2N^-$	*n*-Butyl	0	0	0
[BMI][TF][b]	$CF_3CO_2^-$		0	0	0
[BMI][OTf][b]	$CF_3SO_3^-$		62	0	0
[HMI][PF_6][b]	PF_6^-	*n*-Hexyl	90	3	3
[OMI][PF_6][b]	PF_6^-	*n*-Octyl	82	20	21
CH_2Cl_2	—	—	70	18	19

Compound **1** was extracted from the ionic liquid using:[a] *tert*-butyl methyl ether.
[b] *n*-heptane. [d]diethyl ether.
[c] reaction was performed at 70°C.



Scheme 1. One step deuteration of *N*-(4-methoxyphenyl)-*N*-methyl benzamide 1

H/D Exchange with different catalysts

We briefly examined the generality of [BMI][PF_6] as a solvent for other iridium exchange catalysts by comparing the outcome of labelling **1** with that in CH_2Cl_2, using a selection of mono- and bi-dentate catalysts **2**, **3**, **4**, **5**, and **6**. Results are illustrated in Table 2. Catalyst **5** was prepared by addition of the PPF ligand[§] to [Ir(cod)$(py)_2$]PF_6[8] using a similar method of Bedford *et al.*[9] Catalyst **6** was prepared by first formation of [Ir(cod)$(py)_2$]BARf using a similar method of Crabtree and Morehouse[8]

[§] Ligand was purchased from Solvias AG, Werk Rosental WRO-1055, Schwarzwaldallee 215, CH-4058 Basel, Switzerland.

Table 2. Deuteration of compound 1 with different catalysts in [BMI][PF_6] ionic liquid. Values in brackets ( ) denote reactions performed in CH_2Cl_2

Catalyst	A (%)	B (%)	C (%)
[Ir(cod)[Cy_3P)(py)]PF_6 **2**	63 (99)	19 (36)	20 (71)
[Ir(cod)(dppb)]PF_6 **3**[9]	73 (95)	39 (47)	66 (76)
[Ir(cod)(PPh_3)$_2$]PF_6 **4**[10]	70 (94)	0 (0)	78 (40)
[Ir(cod)(PPF)]PF_6 **5**	81 (95)	90 (95)	90 (98)
[Ir(cod)[Cy_3P)(py)]BARf **6**	78 (96)	2 (64)	2 (96)

followed by addition of PCy_3 to the complex in methanol. All subsequent deuteration reactions were conducted under identical conditions employing 10 mol% catalyst, 1 atm. deuterium gas at rt.

With the exception of **6** (bearing the highly lipophilic [BARf] anion) all other catalysts in [BMI][PF_6] behaved comparably to CH_2Cl_2 with regard to both regiospecificity and net deuterium incorporation. In addition, zero deuterium exchange took place in position B of **1** (3rd entry) in the presence of the monodentate catalyst **4**, as predicted by the Heys model.[11] In general, for lipophilic substrates, incorporation values from ionic liquids were approximately 10% lower than those obtained in dichloromethane. The striking difference between [BMI][PF_6] and CH_2Cl_2 is the rate of reaction. Two experiments were run in parallel, one in CH_2Cl_2 and the other in [BMI][PF_6], employing substrate **1** in the presence of 10 mol% catalyst **2** in each case. Samples were removed at specific time points and deuterium incorporation into **1** was measured by ^{1}H NMR spectroscopy. In CH_2Cl_2, the reaction is complete within 2 h in contrast to 6 h in the ionic liquid. The difference in rates may be due to the higher viscosity of the ionic liquid leading to slower mixing rates and/or reduced solubility of deuterium gas in the more viscous ionic liquid (rate of incorporation vs. stirring rate).[¶] In addition, deactivation of the catalytic species over extended time periods through dimerisation processes may account for the somewhat reduced incorporation.

Application to tritium

To examine for the presence of an isotope effect, compound **1** was stirred in [BMI][PF_6] under 0.9 atm tritium gas at room temperature

[¶] Lower incorporation rates were observed at lower mixing rates, due to lower mass transport of deuterium gas into solution.

with 10 mol% catalyst **5** for 6 h. Work up followed by ^{1}H and ^{3}H NMR analysis revealed 95% tritium incorporated into position A, 95% into B, and 78% into C. Furthermore, analysis of the ionic liquid confirmed no trace of tritiation of [BMI][PF_6].

Application to insoluble substrates

Previous work by Hesk *et al.*[2] revealed that both hydroxyl and carboxylic acid substituted acetanilides **7** and **8** were poor deuteration substrates as a result of their low solubility in dichloromethane (Figure 1). After exposure to 1 atm., deuterium gas, 5 mol% Crabtree catalyst, 16 h, they obtained only 22.5% deuterium labelling into *N*-(4-hydroxyphenyl)acetamide **7** compared with incorporation values ranging between 73 and 96% for more soluble acetanilides. An even lower incorporation rate was obtained for 4-acetylaminobenzoic acid **8**, which under the same conditions, labelled in only 2% overall deuterium incorporation.

In contrast to CH_2Cl_2, both sparingly soluble compounds **7** and **8** dissolved completely in [BMI][PF_6] at room temperature. Several experiments were conducted in this solvent in the presence of both Crabtree catalyst **2** and catalyst **5** at 10 mol% loading. Meanwhile, a second set of reactions were run in CH_2Cl_2 for comparison. The results obtained are illustrated in Table 3. As anticipated, compound **7** deuterated poorly in CH_2Cl_2 (10% overall) due to its low solubility, however, we were pleased to observe much higher deuterium incorporation (58% overall) using [BMI][PF_6] solvent as a result of greater solubility. In the second example, compound **8** failed to be labelled using [BMI][PF_6] in the presence of Crabtree catalyst, however some labelling (5%) was observed using CH_2Cl_2. One may attribute the

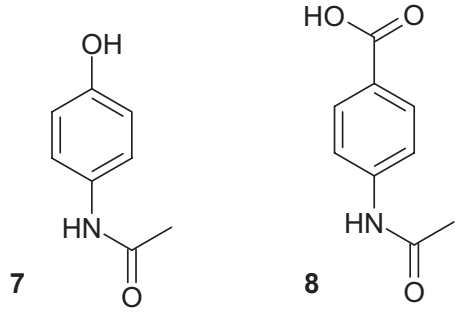


Figure 1. Polar hydroxy- and carboxylic acid acetanilides.

Table 3. Deuteration of compounds 7 and 8 in [BMI][PF_6] ionic liquid at 10 mol% catalyst loading. Values in brackets ( ) denote reactions performed in CH_2Cl_2

Substrate	Catalyst	D incorporation [%]	
		ortho-NHCOMe	*ortho*-COOH
7	**2**	59 (8)	—
	5	57 (12)	—
8	**2**	0 (5)	0 (5)
	5	75 (8)	76 (9)

failure of this reaction to interaction of the free carboxylic acid group with the catalyst giving rise to an inactive coordinatively saturated catalyst species. By selecting catalyst **5**, which exhibits tolerance towards carboxylic acids,[3] we were able to obtain an unprecedented 75.5% overall deuterium incorporation (compared to 8.5% overall in CH_2Cl_2 using the same catalyst) through the combination of both solvent and catalyst selection.

Deuteration of substrates in their salt form

The labelling of salts was briefly investigated with mixed results. Since a number of pharmaceutical drugs are supplied in a salt form for reasons of stability, it would be convenient to label such compounds without prior release of the salt, followed by its subsequent re-conversion. As well as introducing additional steps, this poses stoichiometry challenges on the micro-mol scale.

We examined three different salts, namely, $RCOO^-K^+$, $RNH_3^+Cl^-$ and ArO^-Na^+ as illustrated by the following four compounds, (Figure 2).

Substrates **11**–**13**, potentially good labelling candidates, failed to label even under super stoichiometric amounts of catalyst. One explanation for the apparent lack of activity may be the availability of chloride ions irreversibly binding to the iridium metal centre. Similarly in the case of the phenoxide ion of **13**, strong complexation could lead to complete catalyst deactivation. In our hands only the potassium salt of 2-phenyl-4-quinolinecarboxylic acid **14** labelled (20% at the 3-position) in contrast to the free acid (0%) using 10 mol% Crabtree catalyst, although higher loadings (>110 mol%) may lead to some degree of labelling in the free acid form. In general, we believe that salts are unsuitable candidates for iridium (I) catalysed reactions compared to

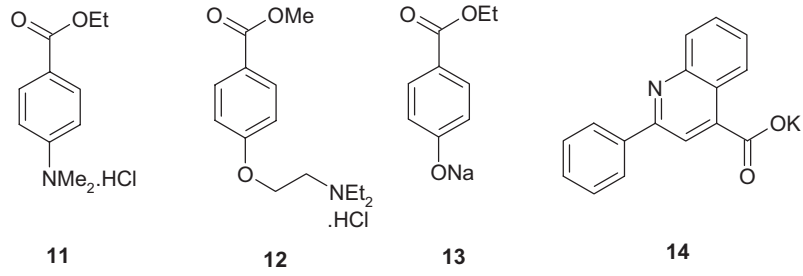
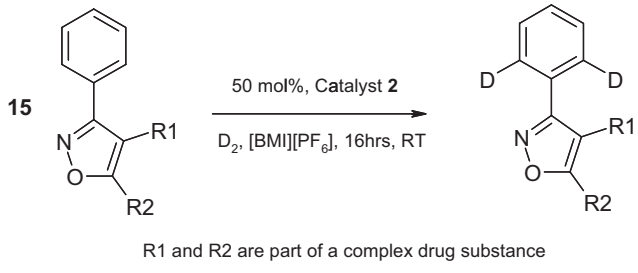


Figure 2. Labelling substrates in their salt forms.



Scheme 2. H/D Exchange labelling of development compound 15

the free acid or base forms since anions such as chloride ions play a crucial part in catalyst deactivation.

Labelling complex molecules

As a final proof of concept, compound **15** (only a partial structure is represented due to patents pending) developed for the treatment of Parkinson's disease, was dissolved in [BMI][PF_6] and exposed to deuterium gas at 1 atmosphere, rt., in the presence of 50 mol% Crabtree catalyst. We were pleased to observe that after 8 h reaction, labelling took place at positions 2- and 6- of the phenyl substituent in 90% overall deuterium incorporation (Scheme 2).

Experimental

All ionic liquids can be synthesised according to literature procedures with the exception of [EMI][NTf_2], [EMI][TF] and [BMI][TF].[‖] Ionic

[‖] Synthesised by unpublished procedures of B. Wietfeld of Novartis Pharma AG.

liquids, [EMI][BF_4], [EMI][PF_6], [BMI][BF_4], [BMI][PF_6], [HMI][PF_6] are commercially available from Aldrich. All catalysts were synthesised from literature procedures with the exception of Crabtree's catalyst **2** purchased from Johnson Matthey plc. All other reagents and solvents were obtained from commercial sources and used without further purification. ^{1}H NMR analysis: Bruker DPX400; $CDCl_3$ or CD_3OD, d_6-DMSO solvents were used. Percentage deuterium incorporation was calculated from ^{1}H NMR spectra. For reasons of reproducibility, all ionic liquids were stored in a glove box (due to the hygroscopicity of some) and removed immediately before use. Separation of *N*-(4-methoxyphenyl)-*N*-methyl benzamide **1** from ionic liquids was achieved by extraction with an appropriate immiscible solvent, either *n*-heptane, *t*-butyl methyl ether or diethyl ether as indicated in Table 1. Separation of polar compounds from [BMI][PF_6] was achieved either by extraction into water or if necessary, first acidification or basification to form a salt followed by extraction into water. All deuterium exchange reactions were performed under 1 atmosphere in a gas tight Schlenk type flask equipped with a magnetic stirrer. All reactions were performed on a 40 μmol scale, employing 0.5 ml ionic liquid and 10 mol% catalyst relative to substrate. Tritiation of **1** was performed using 100% T_2 at 0.9 atm. on a commercial tritium manifold stirring for 6 hours at room temperature. Both product and ionic liquid were analyzed by ^{1}H and ^{3}H NMR spectroscopy.

Conclusion

From the results presented, 1-butyl-3-methylimidazolium hexafluorophosphate supports efficient Ir(I)-catalyzed hydrogen/isotope exchange of poorly soluble substrates, thereby extending the scope of the method and thus reducing the need to invest in costly and time consuming synthetic preparations. Fortuitously, [BMI][PF_6] is commercially available therefore eliminating the need for prior preparation and purity controls. With respect to dichloromethane, catalyst efficacy and regioisotopic distribution of the label is conserved with no undesirable incorporation into the solvent, therefore minimising the risk of contamination. Furthermore, rapid separation of water soluble isotopomers is supported as a result of this ionic liquid's low miscibility with water.

Acknowledgements

We wish to express our gratitude to Dr. Bernhard Wietfeld of Novartis Pharma AG for generously donating ionic liquids for this study and to Prof. Kenneth Seddon from the University of Belfast for helpful discussions.

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