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Metallation of heteroaryls and its application to isotopic labelling

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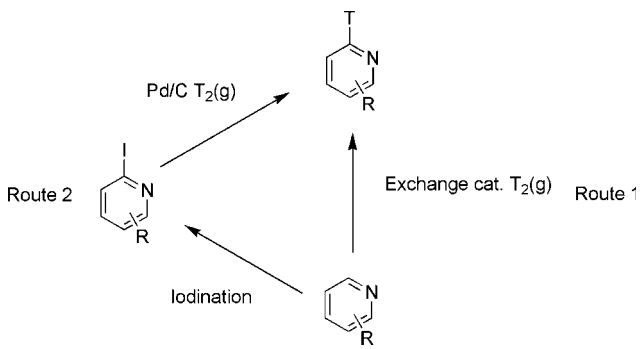
The recently published procedure for the metallation of heteroaromatics and their subsequent reaction with iodine has been applied to the synthesis of deuterium-labelled compounds. The mixed magnesium/lithium base (2) has been prepared and reacted with a range of heteroaromatics. The metallated compounds were iodolyzed with molecular iodine and the resulting iodo compounds were reductively dehalogenated with deuterium gas. This allowed confirmation of the regiochemistry of the metallation and the use of the procedure as a labelling method.

Keywords: tritium; deuterium; iodination; reductive dehalogenation; metallation

Introduction

Rapid access to isotopically labelled molecules is an important aspect of drug discovery and development. Tritium labels can mainly be synthesized from tritium gas using either exchange procedures employing a suitable catalyst, route 1, or via a reductive procedure of a suitable halogenated derivative, route 2, as exemplified by Scheme 1.

Catalysts such as Crabtree's ([Ir.(cod).(PCy_3).(py)]PF_6) are often employed in exchange procedures as they can be both quick and efficient in the incorporation of an isotope into a molecule.[1] However, they do also have drawbacks and can often fail with many complex molecules due to unproductive binding and the need to carry out the exchange reactions in dichloromethane. Of increasing utility is the two-step iodination–reductive dehalogenation procedure which works with a wide variety of activated and deactivated aromatic systems. One potential disadvantage of this approach is the use of strongly acidic conditions e.g. *N*-iodo-succinimide/trifluoroacetic acid[2] to introduce iodine into the molecule. However, once the iodinated derivative has been prepared, it can usually be reduced with either deuterium or tritium gas. This allows the isotope to be incorporated under very mild palladium-catalyzed conditions.

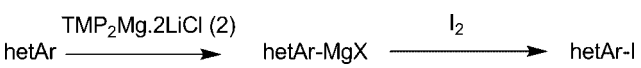


Scheme 1.

The availability of different chemistries to introduce iodine into both aryl and heteroaryl compounds would provide a very important extension to the above approach. One such recently published method involves the metallation of heteroaryls (hetAr) with mixed magnesium/lithium bases such as $TMP_2Mg \cdot 2LiCl$ (**2**; TMP = 2,2,6,6-tetramethylpiperamidyl)[3,4] Scheme 2. The resulting magnesiated species (hetAr-MgX) are then iodolyzed with molecular iodine to form iodo derivatives (hetAr-I). Compound (**2**) has been prepared in our laboratory and used to metallate a range of heteroaryls to demonstrate the utility of this approach. These compounds expand on the known scope of this reaction and provide potential access to new iodo and labelled compounds.

Results and Discussion

The magnesium/lithium base (**2**) can be readily prepared in two steps (Scheme 3) by reacting the lithium chloride salt of isopropylmagnesium chloride with 2,2,6,6-tetramethylpiperidine (TMPH) to form magnesium amide (**1**). This magnesium amide can then be treated with the anion of tetramethylpiperidine (formed by reacting TMPH with BuLi) to form magnesium amide (**2**).[4] Solutions of (**1**) can be stored under nitrogen at ambient



Scheme 2.

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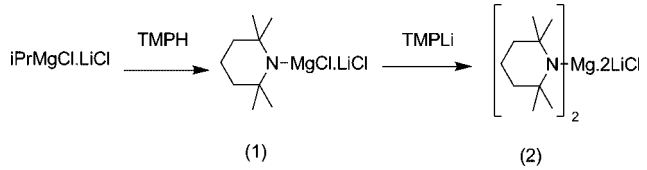
temperatures and used over a period of weeks; however, in our hands solutions of (**2**) need to be used over a period of not more than 1 week before a reduction in quality is observed.

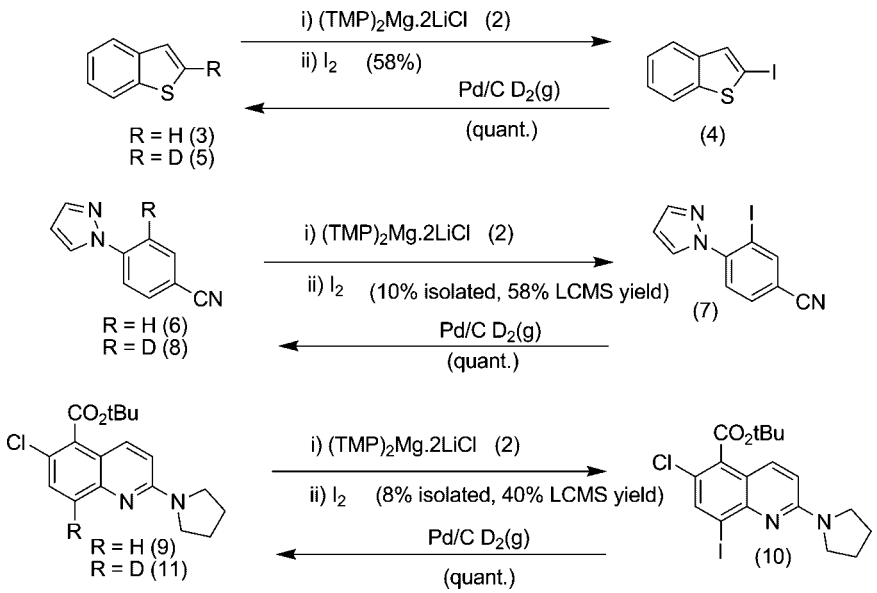
Ultimately we wanted to be able to metallate complex drug molecules with magnesium base (**2**) to allow formation of the iodo and then deutero/tritio versions of the molecule. Initially we started with relatively simple molecules such as benzothiophene (**3**). This was metallated by reaction with (**2**) and on quenching with iodine gave 2-iodo-benzothiophene (**4**) in a reasonable isolated yield (Scheme 4). This iodo compound (**4**) was readily reduced with deuterium gas under mild palladium catalysis to afford deutero-parent (**5**). This allowed confirmation of the regiochemistry of the iodination and the use of the method as a labelling process.

Similarly, the pyrazolylbenzonitrile (**6**) was iodinated in somewhat lower but still synthetically useful yield. The synthesis of iodo (**7**) showed that the magnesium amide was compatible with a cyano group. Nuclear magnetic resonance (NMR) methods (namely n.O.e) allowed the structure to be confirmed. Iodination of (**6**) with NIS/TFA gave an alternative product with the iodine on the pyrazole ring. In fact, other methods were attempted to synthesize iodo (**7**) but were unsuccessful in our hands. The iodo compound (**7**) was then reduced to afford the deutero-parent (**8**) under palladium catalysis with deuterium gas. The more heavily substituted quinoline (**9**) could be metallated and quenched with iodine to allow synthesis of the 8-iodo-quinoline (**10**) in the presence of chloro and *tert*-butyl ester groups. The iodo-quinoline (**10**) was reductively dehalogenated as before to afford the deutero-parent (**11**). This allowed confirmation of the regiochemistry of the iodination. Attempts to metallate the latter two examples with more traditional alkyl lithiums would no doubt lead to complex mixtures due to their reaction with the cyano and ester groups in the molecules. Although the yields from these metallations were low, they were unoptimized and were still synthetically useful for labelling procedures. On obtaining clean, milligram quantities of the iodo compounds, reductive dehalogenation reactions with deuterium and ultimately tritium can easily be carried out.

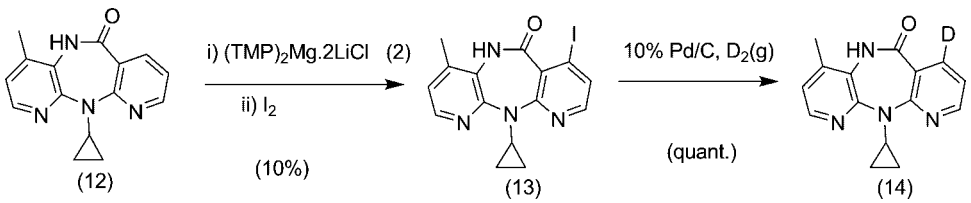
The metallation reaction was then applied to a more complex drug molecule, Neviripine (**12**), an HIV drug, as shown in Scheme 5. Although again the isolated yield was low, the reaction was unoptimized and it is hard to see how the iodo or a high specific activity tritium labelled Neviripine could be synthesized by an alternative method. This is in part due to the compounds poor solubility, which precludes it from reaction with Crabtrees, or Crabtrees type isotope exchange catalysts that require the parent compound to be soluble in dichloromethane. An alternative approach to labelling heteroaromatics[5] using



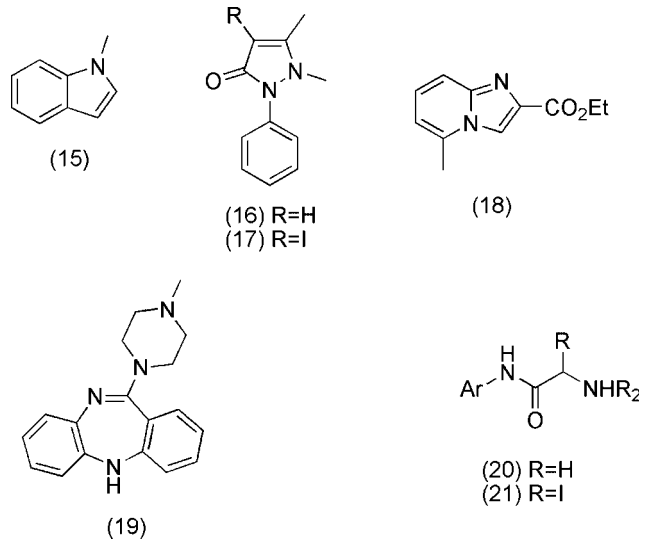
Scheme 3.



Scheme 4.



Scheme 5.



Scheme 6.

rhodium black or ruthenium black catalysts and deuterium or deuterium tritide has been used. However, this only affords low specific activity tritium labels. The iodo Neviripine (**13**) was reduced to the deutero label (**14**) using the conditions described before. This allowed confirmation of the position of the iodo in (**13**) by comparison of the proton NMR of (**14**) with parent (**12**).

We have found some limitations to the use of magnesium amide (**2**) as shown in Scheme 6. The *N*-methyl indole **(15)** does not react at all with (**2**) which might be due to the methyl group blocking any coordination of the magnesium to the indole nitrogen. Antipyrin (**16**), an analgesic drug, is metallated with (**2**) but on quenching with iodine is found to have reacted on the more reactive pyrazolinone ring to give (**17**) rather than on the phenyl ring. This iodo compound (**17**) can be accessed by alternative means.[6] Both the fused pyridyl-imidazolide (**18**) and Clozapine (**19**), a widely used standard in various assays, are unreactive to (**2**). Although the latter bears some structural resemblance to Neviripine (**12**), the lack of ring nitrogen groups or an amide in Clozapine probably precludes it from being able to coordinate to the magnesium base. Finally, a small range of *N*-linked amides (**20**) failed to metallate/iodinate on the heteroaromatic ring presumably due to the presence of an acidic methylene group in the molecule. It was assumed that the iodo compounds (**21**) were formed instead as the iodine largely disappeared from the molecule during aqueous work up. These molecules would be unsuitable precursors to isotopic labels as the incorporated label would be in an exchangeable position.

Experimental

Materials and methods

The synthesis of the magnesium bases (**1**) and (**2**) is described in the original references.[3,4] However, we have found that heat dried, nitrogen flushed standard glassware can be used instead of Schlenk flasks. All reactions were subsequently carried out under a nitrogen atmosphere. All starting materials were purchased from commercial sources and used without further purification except (**9**) and (**20**) that were prepared using literature procedures.[7,8] Standard anhydrous, sure seal THF Fluka cat. ref. 87371 was used without any further drying. Deuterium gas (99 atom%) was purchased from CK Gas Products Ltd. (Hook, Hampshire, UK). NMR spectra were recorded on a Varian Unity Inova 400 MHz NMR spectrometer. Mass spectra (LCMS) were determined on a Waters micromass ZQ with an ESCi probe and Waters Acquity UPLC machine. Gas chromatographic mass spectra (GCMS) were determined on a Hewlett Packard MS 5973 and GC 6890 with electron impact ionization.

General procedure for metallations

A representative example of the metallation, subsequent reaction with iodine and reductive dehalogenation is shown with the synthesis of (**13**) and (**14**).

11-cyclopropyl-7-iodo-4-methyl-5H-dipyrido[3,2-b:2′,3′-e][1,4]diazepin-6(11H)-one (13)

A dry and nitrogen-flushed 10 mL three-necked-flask, equipped with a magnetic stirrer bar and a septum, was charged with 11-cyclopropyl-4-methyl-5H-dipyrido[3,2-b:2′,3′-e][1,4]diazepin-6(11H)-one (**12**) (145 mg, 0.54 mmol) in THF (2.5 ml) and cooled to 0°C. Dilithium magnesium *bis*(2,2,6,6-tetramethylpiperidin-1-ide) dichloride (**2**) (0.707 mL, 0.54 mmol) was added dropwise. After the addition was complete, the reaction mixture was stirred at room temperature for 60 min. The reaction was re-cooled to 0°C and dilithium magnesium *bis*(2,2,6,6-tetramethylpiperidin-1-ide) dichloride (**2**) (0.500 ml, 0.385 mmol) was added. The reaction was allowed to warm to room temperature and stirred for 20 min. The reaction was re-cooled to 0°C and dilithium magnesium *bis*(2,2,6,6-tetramethylpiperidin-1-ide) dichloride (**2**) (0.200 ml, 0.154 mmol) was added. The reaction was allowed to warm to room temperature and stirred for 10 min. N.B. this sequential addition of (**2**) appeared to be critical in driving the reaction towards completion. The reaction was cooled to 0°C and a solution of Iodine (152 mg, 0.60 mmol) in THF (1.5 ml) was slowly added, then allowed to warm to room temperature. The reaction was quenched with sat. ammonium chloride solution, diluted with water and extracted with ethyl acetate. The organic layer was separated, washed with sodium thiosulfate solution (x2), then water, dried ($NaSO_4$), filtered and the solvent removed in *vacuo* to give crude product. This was purified by chromotography on silica (5% ethyl acetate in *i*-hexane rising to 10 then 15% ethyl acetate, 10 g column) the product containing fractions were combined and the solvent removed to give 11-cyclopropyl-7-iodo-4-methyl-5H-dipyrido[3,2-b:2′,3′-e][1,4]diazepin-6(11H)-one (**13**) (18.00 mg, 10%).

LCMS *m*/z 393 ([M+1]+).

[7−^{2}H$_1$]-11-cyclopropyl-4-methyl-5H-dipyrido[3,2-b:2′,3′-e][1,4]diazepin-6(11H)-one (14)

11-cyclopropyl-7-iodo-4-methyl-5H-dipyrido[3,2-b:2′,3′-e][1,4]-diazepin-6(11H)-one (**13**) (18 mg, 0.05 mmol) was dissolved in ethanol (1.5 ml) and 10% palladium on carbon (10 mg, 0.09 mmol) and triethylamine (25 µl, 0.18 mmol) were added. The flask was placed on a deuterium manifold, evacuated and charged with 1 atm of deuterium. The reactants were stirred for 20 h then filtered and the filtrate evaporated to give [7-^{2}H]-11-cyclopropyl-4-methyl-5H-dipyrido[3,2-b:2′,3′-e][1,4]diazepin-6(11H)-one (**14**) (14.00 mg, 114%) as a residue with minor impurities.

^{1}H NMR (d6-DMSO) δ 9.90 (1H, br s), 8.51 (1H, dd), 8.08 (1H, d), 7.20 (1H, dd), 7.07 (1H d), 3.62 (1H, m), 2.34 (3H, s), 0.88 (2H, m), 0.35 (2H, m) which is consistent with the loss

of the 7-proton in comparison to the NMR of the unlabelled parent.

LCMS *m/z* 268 ($[M+1]^+$). The material co-chromatographs with an authentic sample of the protio parent by UPLC.

2-iodobenzo[b]thiophene (4)

2-iodobenzo[b]thiophene (**4**) was prepared from benzo[b]thiophene (**3**) in a manner analogous to the above example (**13**).

^{1}H NMR ($CDCl_3$) δ 7.76 (1H, m), 7.62 (1H, m), 7.52 (1H, s), 7.31 (2H, m).

^{13}C NMR ($CDCl_3$) δ 144.2, 140.5, 133.8, 124.5, 124.4, 122.3, 121.2, 78.4.

GCMS *m/z* 260 [M^+].

[2-^{2}H]-benzo[b]thiophene (5)

[2-^{2}H]-benzo[b]thiophene (**5**) was prepared from 2-iodobenzo-[b]thiophene (**4**) in a manner analogous to the above example (**14**).

^{1}H NMR (CD_3OD) δ 7.90 (1H, m), 7.83 (1H, m), 7.35 (3H, m) which is consistent with the loss of the 2 proton in comparison to the NMR of the protio parent.

3-iodo-4-(1H-pyrazol-1-yl)benzonitrile (7)

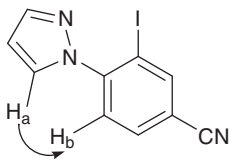
3-iodo-4-(1H-pyrazol-1-yl)benzonitrile (**7**) was prepared from 4-(1H-pyrazol-1-yl)benzonitrile (**6**) in a manner analogous to the above example (**13**).

^{1}H NMR ($CDCl_3$) δ 8.27 (1H, s), 7.87 (1H, d), 7.80 (1H, s), 7.76 (1H, d), 7.55 (1H, d), 6.53 (1H, d).

LCMS *m/z* 296 ($[M+1]^+$).

n.O.e

The n.O.e between pyrazole proton H_a and aromatic proton H_b confirmed the position of the iodine and hence the deuterium in (**8**).



(7)

[3-^{2}H]-4-(1H-pyrazol-1-yl)benzonitrile (8)

[3-^{2}H]-4-(1H-pyrazol-1-yl)benzonitrile (**8**) was prepared from 3-iodo-4-(1H-pyrazol-1-yl)benzonitrile (**7**) in a manner analogous to the above example (**14**).

^{1}H NMR ($CDCl_3$) δ 8.00 (1H, d), 7.84 (1H, d), 7.77 (3H, m), 6.54 (1H, t).

LCMS *m/z* 171 ($[M+1]^+$).

tert-butyl 6-chloro-8-iodo-2-(pyrrolidin-1-yl)quinoline-5-carboxylate (10)

tert-butyl 6-chloro-8-iodo-2-(pyrrolidin-1-yl)quinoline-5-carboxylate (**10**) was prepared from *tert*-butyl 6-chloro-2-(pyrrolidin-1-yl)quinoline-5-carboxylate (**9**) in a manner analogous to the above example (**13**).

^{1}H NMR ($CDCl_3$) δ 8.09 (1H, s), 7.76 (1H, d), 6.75 (1H, d), 3.63 (4H, m), 2.04 (4H, m), 1.64 (9H, s).

LCMS *m/z* 459/461 ($[M+1]^+$).

[8-^{2}H]-*tert*-butyl 6-chloro-2-(pyrrolidin-1-yl)quinoline-5-carboxylate (11)

[8-^{2}H]-*tert*-butyl 6-chloro-2-(pyrrolidin-1-yl)quinoline-5-carboxylate (**11**) was prepared from *tert*-butyl 6-chloro-8-iodo-2-(pyrrolidin-1-yl)quinoline-5-carboxylate (**10**) in a manner analogous to the above example (**14**).

^{1}H NMR ($CDCl_3$) δ 7.82 (1H, d), 7.43 (1H, s), 6.77 (1H, d), 3.61 (4H, m), 2.06 (4H, m), 1.65 (9H, s) which is consistent with the loss of the 8 proton in comparison to the NMR of the protio parent.

LCMS *m/z* 334/336 ($[M+1]^+$).

Conclusion

We have iodinated a number of heteroaromatics with a good range of functionality. Although there are simpler methods for iodinating and labelling molecules, the use of reagents such as (**2**) can be very useful in cases where the molecule of interest has failed to label/iodinate via alternative means. Insolubility of a compound can often be a limitation in the use of exchange catalysts. Generally, solubility problems are not encountered during the metallation reactions as tetrahydrofuran is used as the solvent to carry out the reaction. If the compound is insoluble in THF, a solution will often be obtained as the deprotonation proceeds.

This method also has the potential to access regio-labelled compounds that cannot be synthesized by more traditional labelling or iodinating procedures.

Acknowledgement

I would like to acknowledge Vincent Coissard and Colin Hogg, two excellent students, who did the majority of the synthetic work in this paper. Furthermore, the NMR team at AstraZeneca Charnwood, especially Chris Sleigh and Richard Lewis, have been very helpful with various NMR assignments.

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