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# Synthesis of deuterium-labelled etravirine

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**This study describes the synthesis of deuterium-labelled etravirine. Etravirine labelled with six deuteriums was prepared in eight steps starting from phenol with excellent chemical purity and isotopic enrichment. It provides important internal standard for the clinical studies of etravirine.**

**Keywords:** deuterium; labelled; synthesis; etravirine

## Introduction

Etravirine, a second-generation non-nucleoside reverse transcriptase inhibitor (NNRTI), is highly potent and effective against wild-type and drug-resistant HIV-1 variants. It was granted accelerated approval by the FDA (18 January 2008) and marketed under the name Intelence™. In clinical development, etravirine has shown durable efficacy and, with the exception of rash, a safety profile similar to placebo. Furthermore, the nature and magnitude of adverse events observed with etravirine suggest that the drug may offer improved tolerability over first-generation NNRTIs. In addition it could offer safety benefits over first-generation NNRTIs in early experienced patients. Etravirine can be used in combination with other antiretrovirals such as boosted protease inhibitors and raltegravir with little potential for additional toxicity. Therefore, given this favorable safety profile along with its enhanced barrier to the development of resistance and demonstrated efficacy, etravirine provides an important treatment option for HIV-infected, treatment-experienced patients. The safety of etravirine in treatment-naive patients is being explored in ongoing clinical trials. New trials are also investigating the safety and tolerability of etravirine in treatment-experienced children and adolescents.[1–5] To support the pharmacokinetic and metabolism studies of etravirine, a stable isotope-labelled standard was needed. This study describes the experimental details for the preparation of etravirine labelled with six deuteriums with high chemical purity and isotopic enrichment.

## Experimental

### General

All reagents were obtained from Sigma-Aldrich and CDN Isotope. Mass spectra were recorded using a Quattro micro API mass spectrometer. $^1$H NMR spectra were recorded on a Bruker 300 MHz instrument. Chemical purities were determined by an Agilent 1200 HPLC with a XDB-C18 column, 5 μm, 4.6 × 150 mm.

### Synthesis of (methoxymethoxy)benzene (2)

A 60% dispersion of sodium hydride in oil (15.00 g, 375.0 mmol NaH) was suspended in dry DMF (50 ml) under nitrogen. The cooled (0°C) suspension was slowly treated with a solution of (1) (30.00 g, 318.8 mmol) in dry DMF (50 ml). After the evolution of hydrogen had ceased, the mixture was stirred for 30 min at rt. The cooled (0°C) reaction mixture was treated with chloromethylether (33.00 g, 409.9 mmol). It was stirred for overnight at room temperature, before an ice-cooled saturated $NH_4Cl$ solution (200 ml) was added. The mixture was extracted with diethyl ether (3 × 200 ml). The combined organic layers were washed with 1 M NaOH solution (100 ml), water (200 ml), brine and dried over $Na_2SO_4$. After evaporation of the solvent, the residue was purified by chromatography on silica gel (800 g) column, eluting with petroleum ether, to give (2) as colorless oil (20.00 g, 45.4%).

$^1$H NMR (300 MHz, $CDCl_3$):δ 7.29 (m, 2H), 7.01 (m, 3H), 5.17 (s, 2H), 3.48 (s, 3H).

### Synthesis of 1-(methoxymethoxy)-2-[$^2H_3$]methylbenzene (3)

To a solution of *n*-BuLi in hexane (24 ml 2.2 M, 52.8 mmol) under nitrogen was added slowly TMEDA (6.14 g, 52.8 mmol) at room temperature. The solution was then cooled in an ice bath and (2) (5.00 g, 36.2 mmol) was added dropwise over 15 min. After stirring for 1 h, $CD_3I$ (7.65 g, 52.8 mmol) was added dropwise. After addition, the reaction mixture was allowed to warm to room temperature over 1 h and then quenched with water (20 ml). The solution was extracted with diethyl ether (200 ml). The diethyl ether extracts were washed with 2 M HCl (50 ml), water (3 × 200 ml), brine (200 ml) and dried over $Na_2SO_4$. After concentration, it afforded crude (3) (5.50 g, 97.8%) as a light yellow oil, which was used without further purification.

$^1$H NMR (300 MHz, $CDCl_3$):δ 7.14 (t, $J$ = 6 Hz, 2H), 7.04 (d, $J$ = 6 Hz, 1H), 6.91 (t, $J$ = 9 Hz, 1H), 5.20 (s, 2H), 3.49 (s, 3H).

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### Synthesis of 1-(methoxymethoxy)-2,6-[$^2H_6$]dimethylbenzene (4)

To a solution of *n*-BuLi in hexane (23 ml 2.2 M, 50.6 mmol) under nitrogen was added slowly TMEDA (5.88 g, 50.6 mmol) at room temperature. The solution was then cooled in an ice bath and crude (3) (5.34 g, 34.4 mmol) was added dropwise over 15 min. After stirring for 1 h, $CD_3I$ (7.33 g, 50.6 mmol) was added dropwise. After addition, the reaction mixture was allowed to warm to room temperature over 1 h and then quenched with water (20 ml). The solution was extracted with diethyl ether (200 ml). The diethyl ether extracts were washed with 2 M HCl (50 ml), water (3 × 200 ml), brine (200 ml) and dried over $Na_2SO_4$. After concentration, it afforded crude (4) (5.63 g, 95.0%) as a light yellow oil, which was used without further purification.

### Synthesis of 2,6-[$^2H_6$]dimethylphenol (5)

A solution of crude (4) (5.80 g, 33.7 mmol) in methanol (200 ml) and 2 M HCl (75 ml) was heated at 90°C for 4 h. TLC analysis showed that the reaction was complete. Methanol was removed under reduced pressure, and the residue was extracted with diethyl ether (3 × 100 ml). The combined diethyl ether extracts were washed with water (3 × 100 ml), brine (100 ml) and dried over $Na_2SO_4$. After concentration, the residue was purified by chromatography on silica gel (50 g) column, eluting with petroleum ether, to give (5) (1.26 g, 29.2%) as a colorless crystalline solid.

$^1H$ NMR (300 MHz, $CDCl_3$):δ 6.97 (d, $J$ = 9 Hz, 2H), 6.75 (t, $J$ = 9 Hz, 1H), 4.60 (s, 1H).

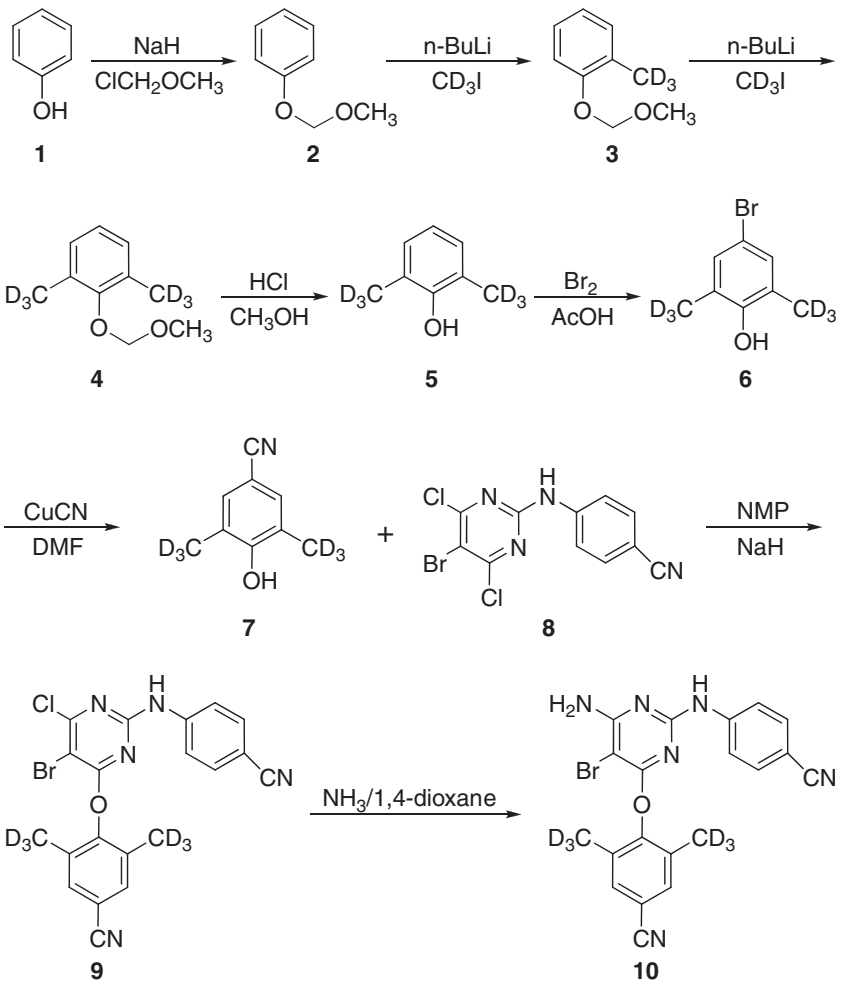
### Synthesis of 4-bromo-2,6-[$^2H_6$]dimethylphenol (6)

To a solution of (5) (0.93 g, 7.3 mmol) in glacial acetic acid (5 ml) cooled in an ice bath was added dropwise a solution of bromine (1.16 g, 7.3 mmol) in glacial acetic acid (5 ml). The reaction mixture was stirred for 20 min at room temperature, and then poured into 10 ml of 1% $NaHCO_3$ solution. The solution was stirred for 10 min and extracted with diethyl ether (3 × 20 ml). The combined organic layers were washed with water (50 ml), brine (50 ml), and dried over $Na_2SO_4$. After concentration, it gave (6) (1.33 g, 88.7%) as off-white solid.

$^1H$ NMR (300 MHz, $CDCl_3$):δ 7.10 (s, 2H), 4.61 (s, 1H).

### Synthesis of 4-hydroxy-3,5-[$^2H_6$]dimethylbenzonitrile (7)

A suspension of (6) (1.33 g, 6.4 mmol) and CuCN (0.66 g, 7.4 mmol) in dry DMF (5 ml) was stirred at 154°C for 5 h. TLC analysis indicated that the reaction was complete. The reaction mixture was cooled to room temperature and then poured into a mixture of $FeCl_3$ (1.60 g), concentrated HCl (0.66 ml) and water (5 ml). The resulting slurry was filtered, washed with water (5 ml) and ethanol (20 ml). The filtrate was concentrated, and then 1 M NaOH solution (50 ml) was added. The resulting mixture was stirred at 50°C for




**Scheme 1.**

30 min. The solid was filtered and washed with water (20 ml). 1 M HCl was added to the filtrate until no more precipitate was formed. The precipitate was filtered and dried over $P_2O_5$ under vacuum overnight to afford (7) (0.57 g, 58.5%) as yellow solid.

$^1$H NMR (300 MHz, $CDCl_3$):δ 7.29 (s, 2H), 5.20 (s, 1H).

### Synthesis of 4-(5-bromo-6-chloro-2-(4-cyanophenylamino)-pyrimidin-4-yloxy)-3,5-[$^2H_6$]dimethylbenzonitrile (9)

To a sealed tube containing a solution of (7) (0.36 g, 2.4 mmol) in 1,4-dioxane (2.2 ml), was added sodium hydride (0.10 g, 2.5 mmol). The mixture was stirred at rt for 2 min. NMP (2.20 g, 22.2 mmol) was added, and the resulting mixture was stirred for an additional 10 min at room temperature. Compound (8) (0.74 g, 2.2 mmol) was added to the mixture, and the vessel was sealed and heated to 155°C for a period of 6 h. After cooling to room temperature, the mixture was diluted with water (15 ml) and the crude product was filtered off and washed with additional water (10 ml). The crude solid was purified by chromatography on silica gel (100 g) column, eluting with $CH_2Cl_2$/Hexane (1:1), to afford (9) (0.83 g, 83.6%) as white solid.

$^1$H NMR (300 MHz, DMSO-$d_6$):δ 10.60 (s, 1H), 7.82 (s, 2H), 7.50 (d, $J$ = 6 Hz, 2H), 7.37 (d, $J$ = 6 Hz, 2H).

### Synthesis of [$^2H_6$]etravirine (10)

Compound (9) (0.41 g, 0.9 mmol) was dissolved in 0.5 M $NH_3$/1,4-dioxane solution(15 ml, 7.5 mmol) in a sealed tube. The vessel was heated to 150°C for a period of 24 h. After cooling to room temperature, the reaction mixture was concentrated *in vacuo*, and purified by chromatography on silica gel (50 g) column, eluting with $CH_2Cl_2$, to afford desired product, which was recrystallized from $CH_3OH$/THF (20 ml, 1:1) to yield (10) (0.16 g, 41.0%) as white solid.

$^1$H NMR (300 MHz, DMSO-$d_6$):δ 9.58 (s, 1H), 7.74 (s, 2H), 7.54 (d, $J$ = 9 Hz, 2H), 7.42 (d, $J$ = 9 Hz, 2H), 7.11 (s, 2H). MS-EI (*m/z*): 439.1 (95), 440.1 (22), 441.1 (100), 442.0 (22). HPLC (XDB-C18, $CH_3OH$/10 mmol/l $CH_3COONH_4$ + 0.03% TEA = 76/24, 1.0 ml/min): $t_R$ 4.8 min (> 97.7%). Isotopic enrichment determined by NMR spectroscopy was over 99.0%.

## Results and discussion

Although there was a synthetic route of [$^2H_6$]etravirine (10) reported by Masse,[6] it was problematic and lacked experimental details. In this patent, the key intermediate, 2,6-[$^2H_6$]dimethylphenol (5) was obtained by treatment of (methoxymethoxy)-benzene (2) with $CD_3I$ and $CD_2I_2$ in sequence. Somehow the use of $CD_2I_2$ caused severe loss of deuterium, and possible gain of deuterium on the aromatic ring.[7]

We decided to prepare (5) by a two-step methylation of (2) with $CD_3I$ followed by hydrolysis.[8,9] As shown in Scheme 1, the hydroxyl group of phenol (1) was protected with chloromethylether to give (methoxymethoxy)benzene (2).[10] Compound (2) was metalated with *n*-BuLi in the presence of *N,N,N′,N′*-tetramethylethylenediamine (TMEDA), followed by methylation with $CD_3I$ to afford 1-(methoxymethoxy)-2-[$^2H_3$]methylbenzene (3).[8,9] In this step, the 1:1 molar complex of *n*-butyllithium with TMEDA in hexane was chosen as the metalating agent due to the higher reaction rates and higher yields in comparison with the use of *n*-butyllithium alone.[11] TMEDA as an additive shifted the reaction from lateral to ortho-lithiation intermediates completely, which avoided the formation of by-products.[9] Treatment of (3) with same procedure produced 1-(methoxymethoxy)-2,6-[$^2H_6$]dimethylbenzene (4). We tried to make (4) from (2) in one step through using excess reagents. It was not successful. The reaction only formed a small amount of compound (4). The major product was compound (3). After standard hydrolysis of (4) with 2 M HCl in methanol, the key intermediate 2,6-[$^2H_6$]dimethylphenol (5) was obtained in over 99% deuterium enrichment and 27% overall yield from (2) over the three steps. Bromination of (5) with bromine in glacial acetic acid gave 4-bromo-2,6-[$^2H_6$]dimethylphenol (6).[12] Treatment of compound (6) with CuCN at reflux in DMF, and then quenching with a solution of $FeCl_3$ generated 4-hydroxy-3,5-[$^2H_6$]dimethylbenzonitrile (7).[12] The reaction of compound (7) and commercially available 4-(5-bromo-4,6-dichloropyrimidin-2-ylamino)benzonitrile (8) in the presence of sodium hydride and 1-methyl-2-pyrrolidinone (NMP) produced 4-(5-bromo-6-chloro-2-(4-cyanophenylamino)pyrimidin-4-yloxy)-3,5-[$^2H_6$]dimethylbenzonitrile (9).[6] Efforts were made to improve the yield for this step through optimizing reaction temperature and time. The best yield of 83% was obtained by heating the reaction at 155°C for 6 h. The ammoniation of compound (9) with 0.5 M $NH_3$/1,4-dioxane in a sealed tube afforded [$^2H_6$]etravirine (10) in 41% yield.[13] It was observed that if $NH_3$ was allowed to leak out of the reaction, a significantly reduced yield was obtained.

After purification by flash chromatography and recrystallization, the desired product (10) was obtained with over 97% chemical purity and over 99% deuterium enrichment, which provided an excellent internal standard for LC-MS-MS studies.

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