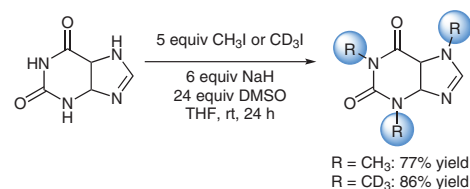# One-Step, Gram-Scale Synthesis of Caffeine-d_9 from Xanthine and CD$_3$I

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Dedicated to Andrew T. B. Stuart, P.Eng., in recognition of his contributions to clean hydrogen and deuterium technologies.

R = CH$_3$: 77% yield
R = CD$_3$: 86% yield

Abstract A one-step, gram-scale synthesis of caffeine-d_9 was achieved using xanthine and CD$_3$I. The reaction proceeds at room temperature using dimsyl sodium as base and THF as solvent, and conducting the reaction on a 1-g scale gave caffeine and caffeine-d_9 in 77% and 86% yield, respectively, after recrystallization.

Caffeine (**1**), or 1,3,7-trimethylxanthine, is a naturally occurring alkaloid found in the leaves, fruit, or nuts of numerous plant species' including coffee, tea, cacao, and so on.[1] It is a psychoactive member of the methylxanthine family of compounds, commonly encountered in food or drinks, where it serves as a mild stimulant, increasing alertness and concentration.[2] In addition to these common uses, it is also considered by the WHO to be an essential medicine, used for the treatment of bronchopulmonary dysplasia and apnea in premature babies.[3] Furthermore, recent studies have suggested caffeine to be effective in treating orthostatic hypotension and hypoxic ischemic encephalopathy,[4] in delaying Alzheimer's progression,[5] and even in inhibiting MCF-7 human breast cancer cells.[6] However, when consumed in excess, acute poisoning can result, with symptoms including nausea, vomiting, insomnia, heart palpitations, and, in rare cases, death.[1,7] Given the ever-increasing consumer consumption of caffeine, especially at higher concentrations, the development of a caffeine alternative that is both long-lasting and equally effective at lower concentrations would have a broad impact.

Caffeine is metabolized *in vivo* through demethylation by the CYP1A2 enzyme, a member of the cytochrome P450 family of enzymes, into its three primary desmethyl metabolites paraxanthine (**3**), theobromine (**4**), and theophylline (**5**) (Figure 1).[8] The pharmacokinetic profiles of caffeine and other compounds metabolized by these enzymes can be modified through deuteration, as substituting hydrogen for deuterium can significantly influence drug-protein binding, receptor affinity, enzyme efficiency, and drug distribution.[1,9] Deuteration can also significantly influence metabolic rates through the kinetic isotope effect, owing to deuterium's increased mass over hydrogen, and the stronger bonds it forms with carbon, which makes such bonds harder to break.[10] Given caffeine's metabolic degradation pathway, it is unsurprising that biological studies on the impact of deuterating its methyl groups (e.g., **1**-d_9) have been conducted over the past few decades.[1,8a,9,11] As caffeine-d_9 re-
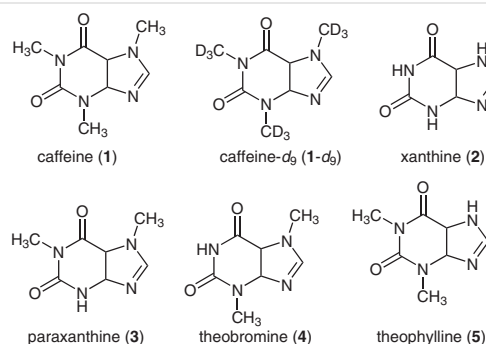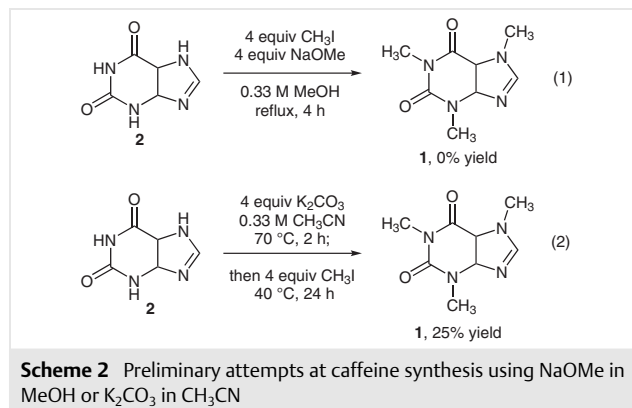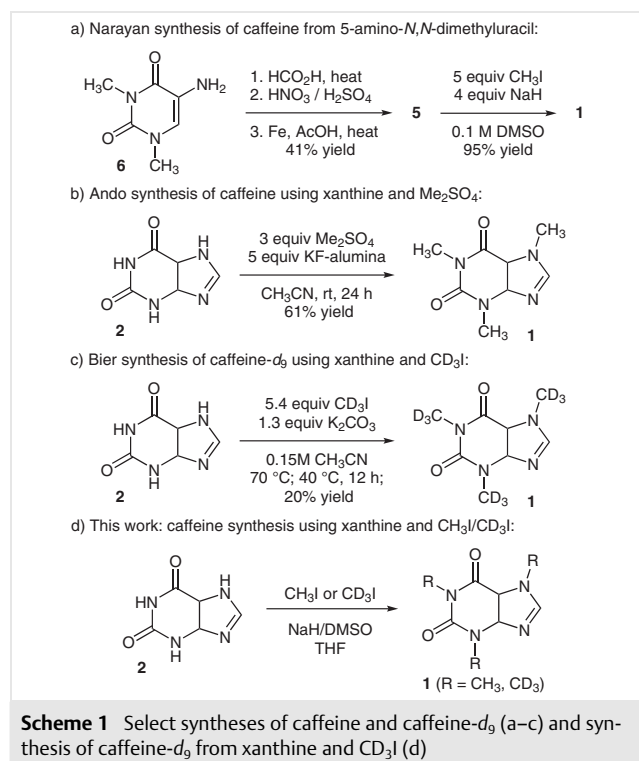

Figure 1 Caffeine (**1**), caffeine-d_9 (**1**-d_9), their parent purine base xanthine (**2**), and their primary desmethyl metabolites paraxanthine (**3**), theobromine (**4**), and theophylline (**5**)

mains an important research target with significant potential for societal and economic impact, devising a cost-efficient, gram-scale synthesis of caffeine-d_9 is warranted.
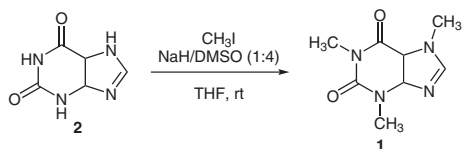
Caffeine is readily obtained as a byproduct of the decaffeinating process, and as there is ample supply of the natural material, little effort has been dedicated to its chemical synthesis. It was first synthesized from uric acid in 1895 by Emil Fischer,[12] but in the intervening century, it received scarce attention from the synthetic community.[13] Syntheses by Traube in 1900,[14] and by Narayan[15] in 2003 followed similar synthetic strategies from 4- or 5-amino-N,N-dimethyluracil, respectively. In the Narayan synthesis, **6** was subjected to formylation, nitration, and reductive heterocyclization steps to give theophylline (**5**), which was then N-methylated to give caffeine (Scheme 1a).[16] In other examples, Ando and co-workers reported a one-step, 61% yield synthesis of caffeine from xanthine using dimethyl sulfate and KF-coated alumina,[16c] and Bier and co-workers published a 20% yielding synthesis of caffeine-d_9 from xanthine using CD$_3$I and K$_2$CO$_3$ (Scheme 1b,c).[17] Unfortunately, much of this precedent is not immediately applicable to a concise, scalable synthesis of caffeine-d_9. For example, existing synthetic routes from natural precursors **3**, **4**, or **5** would not be viable as they already contain non-deuterated methyl groups. A synthesis following the Traube/Narayan strategy would result in significant loss of deuterated material through poor-yielding reactions and purification steps. Ando's use of dimethyl sulfate would not be ideal as dimethyl sulfate-d_6 would be costly to produce and at least half of its

deuterium content would be lost as waste, and Bier's synthesis was operationally challenging and very low yielding. To support the ongoing efforts towards evaluating caffeine-d_9 as an alternative to caffeine, we wished to develop an efficient synthesis of caffeine from xanthine. We report here that a new, one-step synthesis of caffeine from xanthine has been developed, and also that substituting conventional CH$_3$I with CD$_3$I enabled a caffeine-d_9 synthesis without any adverse effect on the reaction outcome (Scheme 1d).

We began our investigation by following the example of González-Calderón's synthesis from theobromine (**4**), and heating xanthine, iodomethane, and sodium methoxide in methanol at reflux (Scheme 2, equation 1).[16d] Unfortunately, no caffeine was observed after 4 h, and extending the reaction time to 24 hours failed to improve on this result. Crude ^{1}H NMR analysis of the reaction mixtures suggested that only mono-methylated xanthine derivatives had been produced. The lack of reactivity in these reactions was attributed to the poor solubility of **2** in both water and alcoholic solvents.[18] We also attempted the reaction in acetonitrile with K$_2$CO$_3$, modifying the procedure of Bier;[17] however, our eventual 25% yield was little better than their reported 20% yield (Scheme 2, equation 2).

Scheme 2 Preliminary attempts at caffeine synthesis using NaOMe in MeOH or K$_2$CO$_3$ in CH$_3$CN

We next consulted the Narayan synthesis from theophylline (**5**),[15] which used 5 equiv of CH$_3$I to achieve a single methylation, and also used the very hazardous base dimsyl sodium.[19] But given the improved solubility of **2** in DMSO, the ability to moderate the risks of NaH/DMSO by diluting with THF,[20,21] and given the significant room for improving the CH$_3$I stoichiometry, we next studied these reaction conditions. We were surprised to find that reacting xanthine with CH$_3$I (5 equiv), NaH (4 equiv), and DMSO (16 equiv) in THF at room temperature gave 45% yield of caffeine after 24 hours (Table 1, entry 1). While increasing the loading of CH$_3$I to 7 equiv resulted in a 53% yield of caffeine (entry 2), this modification was not adopted as we believed it to constitute a poor outcome given the 40% increased loading of methylating agent. Conversely, increasing the loading of NaH in DMSO to 6 equiv gave caffeine in 75% yield, and further increasing it to 8 equiv gave caffeine in a similar 78%

Scheme 1 Select syntheses of caffeine and caffeine-d_9 (a–c) and synthesis of caffeine-d_9 from xanthine and CD$_3$I (d)

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Table 1 Optimization of Caffeine Synthesis Using NaH/DMSO and CH₃I[a]

| Entry	CH$_3$I (equiv)	NaH (equiv)	DMSO (equiv)	Time (h)	Yield (%)
1	5	4	16	24	45
2	7	4	16	24	53
3	5	6	24	24	75
4	5	8	32	24	78
5	5	8	0	24	0
6	5	6	24	18	46
7	5	6	24	48	64
8	4	6	24	24	50
9[b]	5	6	24	24	77

[a] Reactions conducted on a 1 mmol scale in THF (7 × volume of DMSO) at rt. **1** was recrystallized from EtOH.
[b] Reaction conducted on a 1 g scale.

yield (entries 3 and 4). Conversely, using NaH in THF in the absence of DMSO caused the reaction to fail completely (entry 5). Varying the reaction time also failed to improve the yield, as stopping the reaction after 18 hours gave **1** in 46% yield and stopping it after 48 hours gave **1** in 64% yield (entries 6 and 7). At this stage we also attempted to decrease the loading of CH₃I to 4 equiv, but as this only gave caffeine in 50% yield (entry 8), it appeared that the 5 equiv were required to achieve a good yield. Finally, we conducted the reaction on a 1 g (6.6 mmol) scale, to potentially overcome any material losses occurring during the small-scale recrys-

tallization. To our delight, this reaction proceeded without incident, and **1** was recovered in 77% yield (entry 9), in excellent accord with the earlier results.

Having developed an effective, one-step synthesis of caffeine from xanthine, we then attempted the synthesis using CD₃I as the methylating agent. When conducted on a 1 g (6.6 mmol) scale, the reaction proceeded without incident to produce caffeine-d_9 in 86% isolated yield after recrystallization (Scheme 3).[21] Comparison of the ^{1}H NMR spectra for caffeine and caffeine-d_9 showed the expected disappearance of the three methyl peaks (δ = 3.94, 3.51, and 3.34), consistent with their deuteration (Figure 2). Furthermore, while the peak for the C8 proton was still present, it was shifted 0.05 ppm upfield (δ = 7.42 vs 7.47) for deuterated caffeine, consistent with long-range shielding by the N7–CD₃ group. Comparison of the ^{13}C NMR spectra for caffeine and caffeine-d_9 also showed the expected changes consistent with deuteration (Figure 3). While the five carbon signals from the purine core were relatively unchanged, the signals associated with the methyl groups were markedly different. These were observed as septets ($2nI + 1$, where $I_D = 1$ and $n = 3$) with 21.6 Hz coupling constants. Furthermore, their heights were significantly depressed due to the lack of decoupling-based NOE enhancement, and they were shifted 0.8 ppm upfield, again due to the shielding effect of deuterium.[22]

Scheme 3 Synthesis of caffeine-d_9
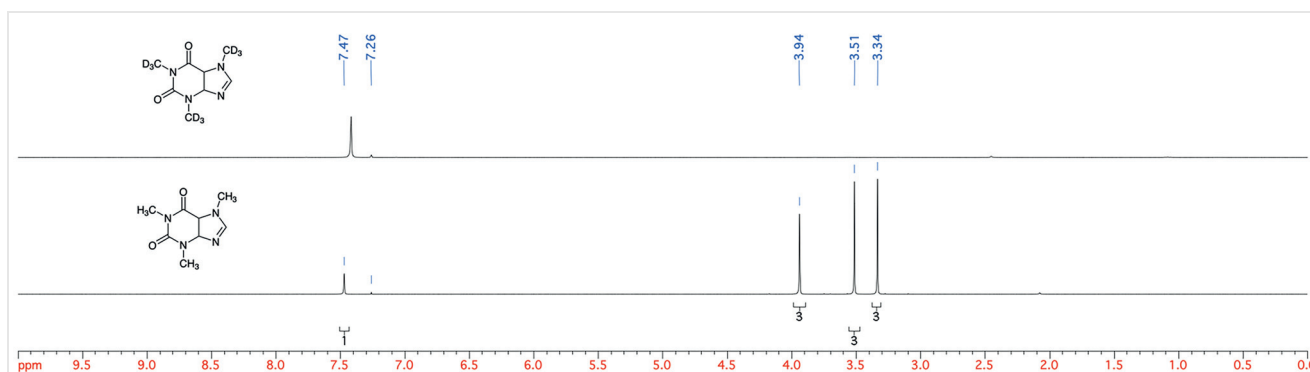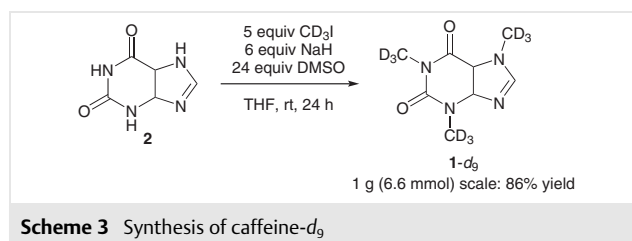
Figure 2 Overlaid ^{1}H NMR spectra of caffeine (bottom) and caffeine-d_9 (top)

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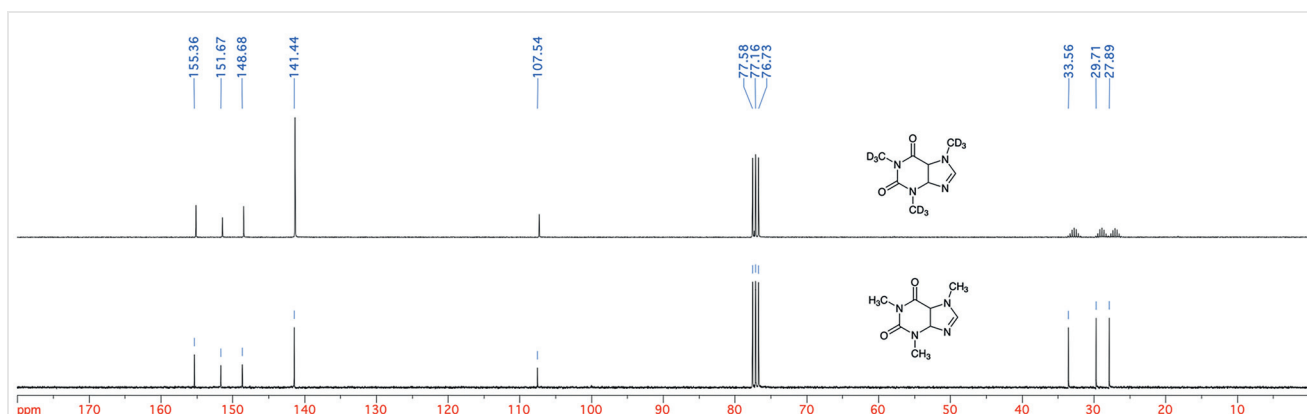
Figure 3 Overlaid ^{13}C NMR spectra of caffeine (bottom) and caffeine-d_9 (top)

In conclusion, we have developed an efficient, gram-scale synthesis of caffeine and caffeine-d_9 from xanthine. Many of the various conditions reported in the literature for methylating xanthine and other desmethyl caffeine precursors were tested, and optimal reaction conditions were found to be a 1.6-fold excess of the methylating agent and using dimsyl sodium as the base. Additionally, the use of THF as the solvent was critical to mitigating the hazards associated with thermal decomposition of the NaH/DMSO mixture. This synthetic route offers a significant improvement over existing strategies for preparing caffeine-d_9, and we anticipate that this route will be benefit researchers seeking gram-scale access to deuterated caffeine and its related derivatives.

Reactions were carried out in oven-dried glassware and cooled under a nitrogen atmosphere. THF was dried and purified using a JC Meyer solvent purification system and was used without further purification. Transfer of anhydrous solvents and reagents was accomplished with oven-dried needles. ^{1}H NMR spectra were recorded at 300 MHz and are reported relative to the residual solvent peak (δ = 7.26). ^{13}C NMR were recorded at 75 MHz and are reported relative to the center line of the residual solvent peak (δ = 77.16). All literature known compounds matched the spectral data found in the literature. HRMS was performed on a Thermo Fisher Scientific Q-Exactive hybrid mass spectrometer. Accurate mass determinations were performed at a mass resolution of 70,000. Samples were infused at 10 µL/min in CH_3OH/H_2O (1:1) + 0.1% formic acid. Xanthine was purchased from Oakwood Chemical and CD_3I was obtained from deutraMed Inc (www.deutramed.com).

Caffeine-d_9 (1-d_9)

To a flame-dried round bottom flask was added THF (78.5 mL) followed by anhyd DMSO (11.2 mL, 158 mmol, 24 equiv). The solution was cooled to 0 °C in an ice bath and NaH (1.58 g, 60% in mineral oil, 39.5 mmol, 6 equiv) was slowly added (300 mg/min), after which the resulting grey suspension was stirred at 0 °C for 30 min. The reaction mixture warmed to rt, xanthine (1 g, 6.6 mmol, 1 equiv) was added and the resulting was mixture stirred for 30 min. CD_3I (2.09 mL, 32.9

mmol, 5 equiv) was then added dropwise over ~5 min and the reaction was then stirred for 24 h at rt. Water (100 mL) was added to the reaction and the resulting biphasic mixture was extracted with DCM (3 × 70 mL). The combined organic phases were concentrated under vacuum to give a crude yellow solid, and this was purified by recrystallization (EtOH) to yield fine, white, fiber-like crystals. The crystals were isolated by vacuum filtration, and the resulting filtrate could be concentrated and subject to further recrystallization. The combined crystals were dried under high vacuum to yield caffeine-d_9 (1.154 g, 86%). NMR and melting point data matched that found in the literature;[17] mp 235–237 °C (uncorrected; Lit.[17] 236.1 °C).

^{1}H NMR (CDCl$_3$, 300 MHz): δ = 7.42 (singlet).

^{13}C NMR (CDCl$_3$, 75 MHz): δ = 155.1, 151.5, 148.5, 141.3 (CH), 107.3, 32.8 (septet, J = 21.6 Hz, CD$_3$), 28.9 (septet, J = 21.6 Hz, CD$_3$), 27.1 (septet, J = 21.6 Hz, CD$_3$).

Conflict of Interest

The authors declare no conflict of interest.

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