Continuous Flow Photochemistry for the Rapid and Selective Synthesis of 2'-Deoxy and 2',3'-Dideoxynucleosides

Continuous Flow Photochemistry for the Rapid and Selective Synthesis of 2′-Deoxy and 2′,3′-Dideoxynucleosides

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A new photochemical flow reactor has been developed for the photo-induced electron-transfer deoxygenation reaction to produce 2'-deoxy and 2',3'-dideoxynucleosides. The continuous flow format significantly improved both the efficiency and selectivity of the reaction, with the streamlined multi-step sequence directly furnishing the highly desired unprotected deoxynucleosides.

Deoxynucleosides have demonstrated prominent antiviral and antitumoral activities,[1] leading to the development of several drugs, including but not limited to zidovudine, stavudine, trifluridine, idoxuridine, cladribine and didanosine (Fig. 1). Extraction from natural sources and fermentation processes can only provide a limited number of naturally-occurring 2'-deoxynucleosides; therefore, synthetic approaches to the preparation of deoxynucleosides are highly desired.[2] The most common approach involves the direct S_N2 reaction of metal salts or silylated nitrogenous bases with 1-chloro-2-deoxyribose derivatives (Scheme 1, Approach A). However, 1-chloro sugars are expensive and unstable, and the substitution reaction often generates a mixture of anomers that are difficult to separate.[2a] Another method involves the radical deoxygenation reaction of ribonucleosides when the C2'-hydroxyl group is derivatized as a phenoxythiocarbonyl ester.[3] Nevertheless, this method is hampered by the use of a stoichiometric amount of toxic reagent $^{n}Bu_3SnH$, and is only applicable to naturally-occurring ribonucleosides.

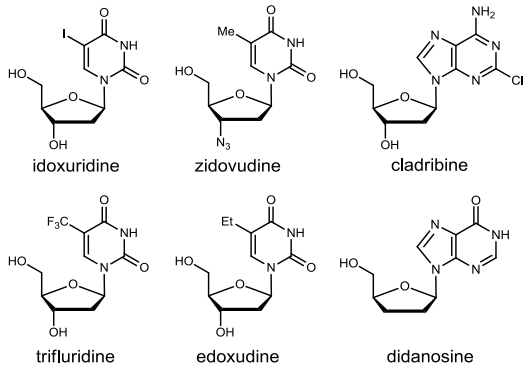
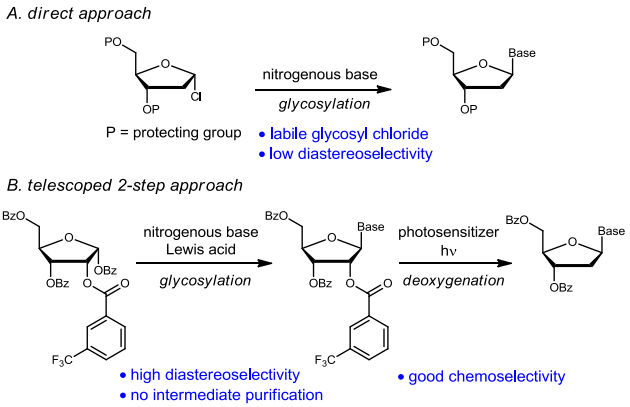


Fig. 1 Selected Deoxynucleoside Drugs

Rizzo and co-workers developed the 2-step *de novo* synthesis of 2'-deoxynucleosides involving Lewis acid mediated Vorbrüggen glycosylation[4] followed by selective C2'-deoxygenation (Scheme 1, Approach B).[5] The C2'-hydroxyl group was derivatized as *m*-CF_3-benzoate, which served as the directing group for the glycosylation. This methodology allows the installation of both natural and unnatural nitrogenous bases with high diastereoselectivity favoring the β-anomer. The 2'-*m*-CF_3-benzoate moiety was then selectively removed (deoxygenated) under photo-induced electron-transfer (PET) conditions.[6] However, the photochemical step required relatively long irradiation time (1.5-2.0 h), and the over-deoxygenation on 3'-position was often observed, leading to diminished yields of the desired 2'-deoxynucleosides.[5c,d]



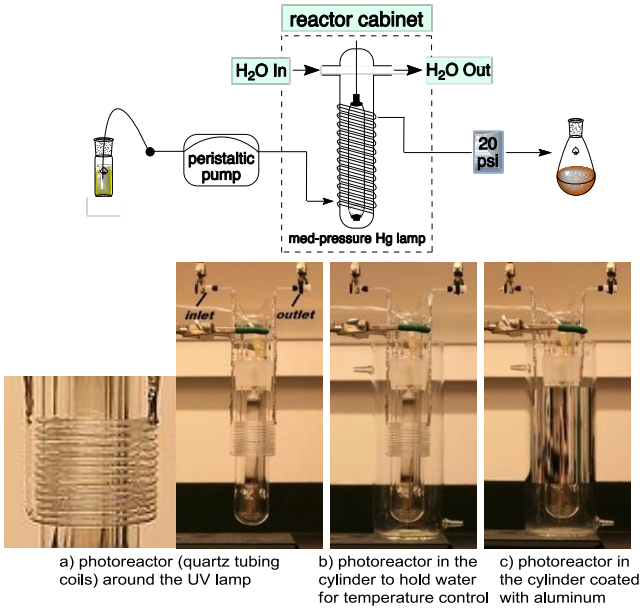
Scheme 1. Approaches toward 2'-Deoxynucleosides

Immersion well reactors are commonly used for photochemical reactions utilizing UV light irradiation in batch. Light intensity significantly decreases as light travels through the reaction solution (Beer-Lambert Law); therefore, only the area close to the UV lamp absorbs enough light and the reaction often takes a long time. When reaction scales change, poor reproducibility is often encountered given the fact that photochemical reactions are strongly dependent on the area of solution exposed to irradiation. Additionally, due to the fixed reactor volume, photochemical reactions are often very challenging to scale up. Continuous flow chemistry has recently emerged as a promising technology,[7] and demonstrated to be an efficient tool for circumventing some of the limitations of traditional batch photochemistry.[8] Larger scales can be achieved by allowing more material to flow for longer periods of time (scale out). The thin channel within the

photochemical reactor allows maximal light transmission and thus significantly increases reactivity. In addition, since the product is continuously removed from irradiation, over reaction and/or detrimental side-reactions can be minimized.

We have recently developed a unique photochemical flow reactor featuring quartz tubing, an aluminum mirror and temperature control, and used this set-up for the efficient synthesis of 2'-deoxy and 2',3'-dideoxy-nucleosides in continuous flow in high yields with good reproducibility.[9] In this account, we disclose more details of our development and the application of our flow photochemical set-up for the reaction mechanism study as well as the multi-step synthesis of biologically interesting targets.

We used a 450 W medium pressure Hg lamp with a Pyrex sleeve (280 nm cutoff) that was positioned in the center of a quartz jacketed immersion well with tap water running through to prevent overheating. Customized coils of quartz tubing (1 mm i.d., 1.84 mL total volume) were placed around the immersion well approximately 2 cm from the surface of the lamp (Fig. 2a). The quartz tubing was extended with PFA tubing, with the entry connected to a peristaltic pump, and the exit connected to a 20 psi back pressure regulator and then a collection vial. The apparatus described above was placed in a Pyrex cylinder through which water was circulated with the aid of a water pump immersed in a temperature controlled bath. The photoreactor (quartz tubing loops) was submerged in the water flowing through the cylinder (Fig. 2b), allowing the reaction temperature to be controlled from 0 to 50 °C by the bath.



a) photoreactor (quartz tubing coils) around the UV lamp

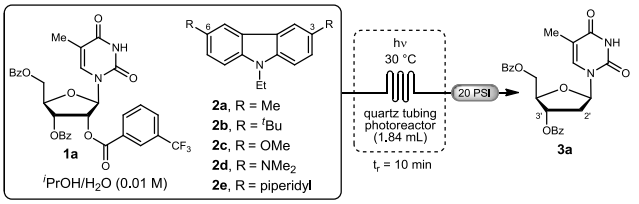
b) photoreactor in the cylinder to hold water for temperature control

c) photoreactor in the cylinder coated with aluminum

Fig 2. Photochemical Flow Reactor Featuring Quartz Tubing and an Aluminum Mirror

9-Methylcarbazole was originally used as a photosensitizer for the PET deoxygenation reaction.[6] 3,6-Dimethyl-9-ethyl-carbazole (**2a**) was later discovered as a more robust photosensitizer which could be used in a catalytic amount.[5c,d] With our flow system, we found that 20 mol% photosensitizer carbazole **2a** catalyzed the PET deoxygenation reaction of *m*-CF_3-benzoate **1a** in ^{i}PrOH/H_2O (9:1) to deliver protected thymidine **3a** in 41% yield in only 10 min at 30 °C. An aluminium-coated cylinder (mirror diameter = 11 cm, Fig. 2c) was employed to increase the light intensity by utilizing the reflection of UV light,[10] and enabled an increase of the yield to 66%. At this stage, several new carbazoles were prepared and examined as photosensitizers (Table 1). 3,6-Dimethoxy-9-ethylcarbazole (**2c**) proved to be the best, affording **3a** in 73% yield (Table 1, entry 4). This is likely attributed to the fact that the more electron-rich substituents on 3- and 6-positions of the carbazole could better stabilize the proposed radical cation intermediate.[11] Unfortunately, more electron-rich carba-zoles **2d** and **2e** bearing the amino substituents decomposed when subjected to UV light (Table 1, entries 5 & 6). When the reaction temperature was raised to 45 °C, only 10 mol% carbazole **2c** was needed to provide **3a** in 84% yield (Table 1, entry 7). The additive $Mg(ClO_4)_2$ used in previous methods[5,6] was not necessary in our system. Indeed, our unique photochemical set-up substantially reduced the reaction time from 2 h in batch to 10 min in flow. More significantly, performing the reaction in the continuous flow fashion suppressed over deoxygenation, namely, the 2',3'-dideoxy derivative was limited to only 2%, while it has been reported as 16% in batch. [5c]

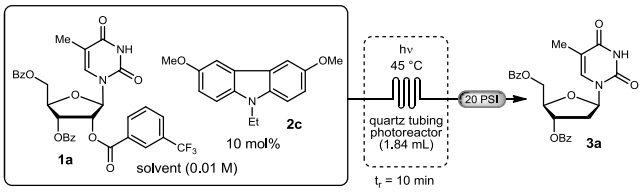
Table 1. Screening of the Photosensitizers



entry[a]	photosensitizer	yield [%][b]
1	**2a**	41
2[c]	**2a**	66
3[c]	**2b**	49
4[c]	**2c**	73
5[c]	**2d**	0[d]
6[c]	**2e**	0[d]
7[c,e]	**2c**	84

a) All reactions were carried out at 30 °C using 20 mol% photosensitizers unless otherwise noted. b) Isolated yield. c) Aluminum mirror was used. d) Carbazole decomposed. e) Reaction was carried out at 45 °C using 10 mol% photosensitizer.

Table 2. Screening of the Solvents

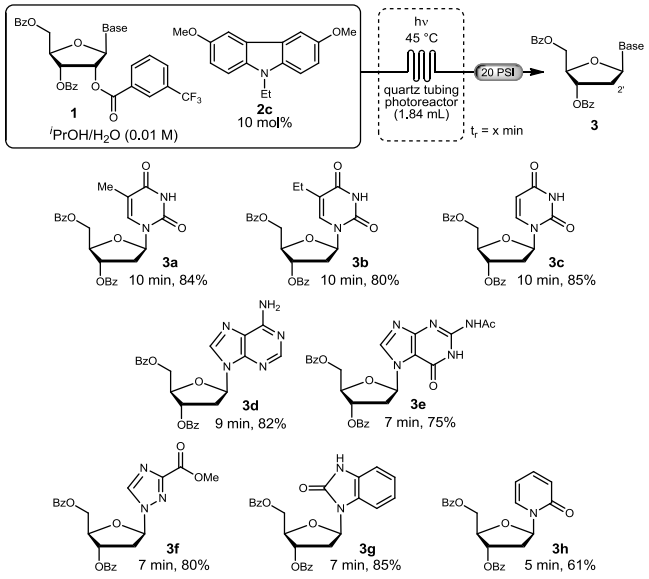


entry	solvent	yield [%][a]
1	MeOH/H_2O (9:1)	18
2	EtOH/H_2O (9:1)	63
3	^{i}PrOH/H_2O (9:1)	84
4	^{i}PrOH	76
5	$CH_2(OMe)_2$	<5
6	$CH(OMe)_3$	<5

a) Isolated yields

A mixture of ^{i}PrOH and H_2O (9:1) has been used as the solvent in Rizzo's studies. Not only as a solvent compo-

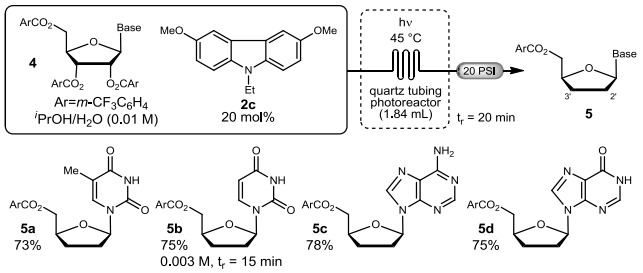
nent, ^{i}PrOH was also proposed to be a hydrogen radical donor for the deoxygenation reaction. We also evaluated other solvent systems (Table 1). Not surprisingly, MeOH/H_2O and EtOH/H_2O generated lower yields (Table 1, entries 1-2), as they are weaker hydrogen radical donors. Isopropanol solely (without water) as a solvent also provided the desired product, although in a slightly lower yield. We envisioned that $CH_2(OMe)_2$ and $CH(OMe)_3$ are better hydrogen radical donors as they would form more stable radicals after hydrogen atom abstraction. However, they only delivered a trace amount of the deoxygenation product, suggesting that a protic solvent is essential for the reaction to proceed (vide infra for mechanistic discussion).



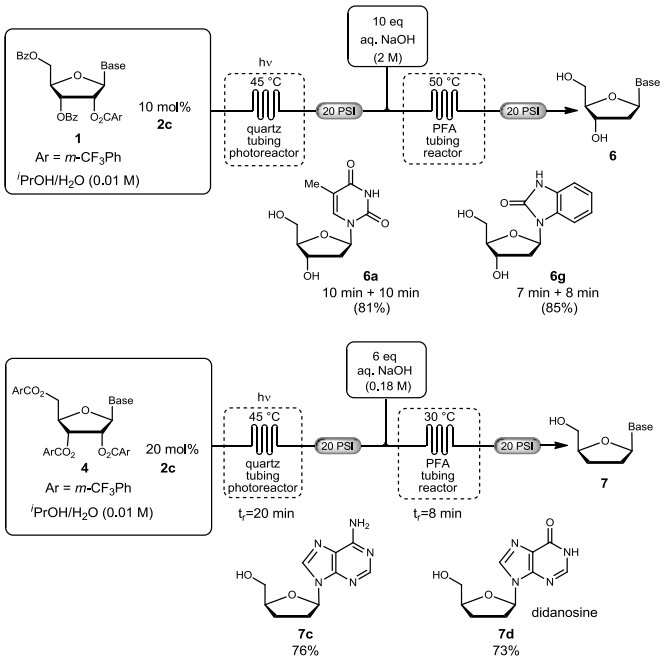
Scheme 2. Synthesis of 2'-Deoxynucleosides in Flow

We investigated the scope of the deoxygenation reaction by surveying a collection of ribonucleosides (Scheme 2). Taking the advantage of the short reaction time under the flow conditions, we were able to rapidly optimize the residence time for each substrate by changing flow rates to maximize yields. Besides **1a**, benzoyl-protected uridine (**1c**), adenosine (**1d**), *N*7-guanosine (**1e**) all generated the corresponding 2'-deoxynucleosides (**3c**-**3e**) in high yields in no more than 10 min residence time.[12] Non-natural nucleosides containing the ethyluracil (**1b**), triazole (**1f**), imidazolone (**1g**) and pyridinone (**1h**) moieties were readily prepared by glycosylation reaction of the protected ribose with the corresponding nitrogenous bases, and they were also successfully deoxygenated with high efficiency. Over 80% yields were obtained in most cases, and the dideoxygenation by-products were often barely detectable. The relatively low yield obtained in the case of **3h** was due to partial decomposition, which was minimized by reducing the residence time to 5 min. Notably, compound **3b** is the precursor to the antiviral drug edoxudine, and compound **3c** is the precursor to the antiviral drug idoxuridine. Reactions in flow were found to be highly reproducible regardless of the scale in all cases.

We also examined the double deoxygenation reaction of triesters **4**, prepared directly from the corresponding nucleosides. Differentiation of the 2'- and 3'-positions is unnecessary in these cases. By increasing the catalyst (**2c**) loading to 20 mol% and extending the residence time to 20 min, the deoxygenation reaction afforded 2',3'-dideoxynucleosides **5a-d** selectively in high yields (Scheme 3). No 5'-deoxy derivative was observed in any case examined. Compound **5d** is of particular interest, as it is the precursor to the anti-HIV drug didanosine.

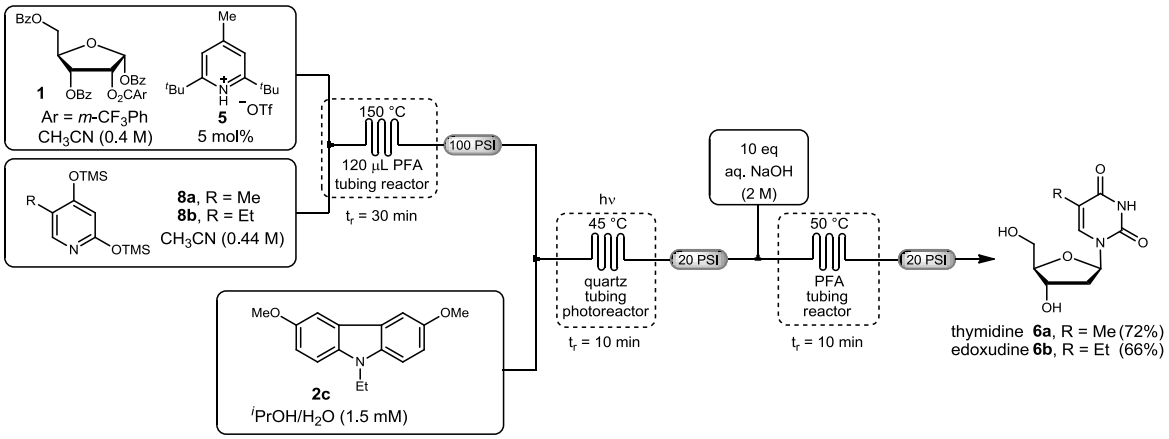


Scheme 3. Synthesis of 2',3'-Dideoxynucleosides in Flow



Scheme 4. One-flow Two-step Synthesis of Deoxynucleosides

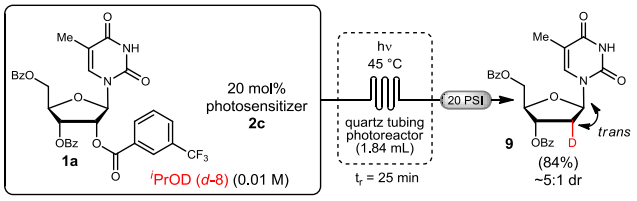
Multi-step synthesis in flow has emerged as a very effective strategy that saves cost and labor by circumventing the need for purifying and isolating intermediate products.[13] However, the development of multistep continuous-flow syntheses remains challenging, as flow-rate synergy, solvent compatibility, and the effect of by-products and impurities must be considered and optimized in downstream reactions. We have demonstrated that an aqueous solution of NaOH could be introduced via a T-mixer to the exiting stream from the PET deoxygenation reaction, and the deprotection occurred in 8-10 min. The photosensitizer and the acid by-product from the first step did not affect the second step. Both fully unprotected 2'-deoxynucleosides and 2',3'-dideoxynucleosides could be prepared using the 2-step-in-1-flow protocol in a very short time and high yields, including the anti-HIV drug didanosine (**7d**) (Scheme 4).



Scheme 5. 3-step-in-one-flow Synthesis of 2'-Deoxynucleosides

We have recently developed a novel Brønsted acid-catalyzed glycosylation reaction for the synthesis of ribonucleosides.[14] This method precludes the use of a stoichiometric amount or an excess of Lewis acids (most commonly $SnCl_4$), and has demonstrated to be a greener and more efficient process. Using this method, we prepared compounds **1a** and **1b** and directly used them for PET deoxygenation without purification in one flow (Scheme 5). The use of CH_3CN as the solvent in the glycosylation step retarded the deoxygenation, presumably due to its absorption of UV light. Therefore, more photosensitizer (15 mol%) was needed to maintain the reactivity. The deoxygenation reaction was immediately followed by the deprotection step. Thus, the 3-step-in-1-flow protocol (glycosylation-deoxygenation-deprotection) successfully afforded thymidine (**6a**) and the anti-HIV drug edoxudine (**6b**) in 72% and 66% yield, respectively. With the total residence time less than 1 hour, this telescoped procedure has demonstrated a more efficient process for the synthesis of 2'-deoxynucleosides.

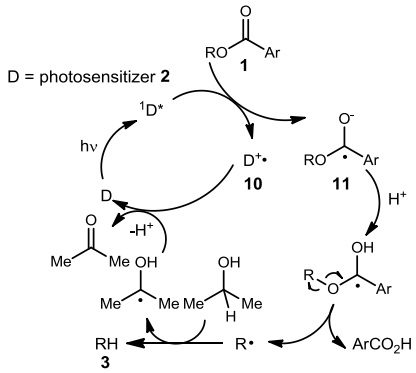
Deuterium-labeled deoxynucleosides are useful for the investigation of interactions of a sugar moiety in DNA with a protein or a drug by NMR spectroscopy.[15] When we used ^{i}PrOD (*d*-8) as the solvent, 2'-deoxy-[2'-D]ribonucleoside **9** was isolated in a good yield with approximately 5:1 dr at C-2', favoring the C1'-C2' *trans* diastereomer (Scheme 6). Similar substrate-controlled diastereoselectivity has been observed in radical deoxygenation[3b, 16] or dehalogenation[17] using Bu_3SnD; the method described herein eschews this toxic and expensive reagent. Deuterium atom transfer was substrantially slower than hydrogen atom transfer. The reaction in ^{i}PrOD-d_8 required the use of 20 mol% carbazole **2a** and 25 min to achieve complete conversion (cf. 10 mol% catalyst and 10 min in ^{i}PrOH). Our flow photochemical set-up not only facilitates the straightforward synthesis of deuterated deoxynucleoside **9**, but also provides a convenient means to investigate the mechanism of the deoxygenation reaction using only a small amount of the substrate and solvent. When $(CH_3)_2CHOD$ was used as the solvent, no deuterium incorporation was identified in the product, and no kinetic isotope effect was observed, clearly indicating that either a CH_3 or CH group (likely the latter) of ^{i}PrOH is the source of the hydrogen atom transferred to the product.



No Deuterium incorporation when $(CH_3)_2CHOD$ was used

Scheme 6. Synthesis of a Deuterium-labeled 2'-Deoxynucleoside

The PET deoxygenation mechanism has been discussed in earlier reports (Scheme 7).[5c,6] Our studies provided further understanding of the reaction mechanism in that: 1) A more electron-rich carbazole photosensitizer facilitates the reaction as it can better stabilize radical cation intermediate **10**; 2) A protic solvent is essential, as radical anion intermediate **11** needs to be protonated to eliminate a carboxylic acid rather than a carboxylate; 3) The methine CH is most likely the source of hydrogen that results in the deoxygenated product.



Scheme 7. Proposed Reaction Mechanism

In conclusion, we have developed a unique flow photochemical reactor featuring quartz tubing, an aluminum mirror, and temperature control. With this setup, PET deoxygenation reactions in flow afforded protected deoxynucleosides in high yields and selectivity, in a short residence time. The new electron-rich carbazole **2c** further improves reactivity. Both natural and non-natural deoxynucleosides

were prepared with high efficiency and reproducibility. The continuous multi-step, one-flow sequence produced unprotected 2'-deoxy and 2',3'-dideoxy ribonucleosides in a streamlined manner, including important drugs edoxudine and didanosine. Our studies also provided further understanding of the reaction mechanism. We believe that this contribution not only improves the synthesis of deoxynucleosides, but will also incite further interest and development of other photochemical transformations in flow.

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