An Environmentally Responsible Synthesis of the SARS-CoV-2 M^{pro} Inhibitor Nirmatrelvir (PF-07321332), the Active Ingredient in Paxlovid

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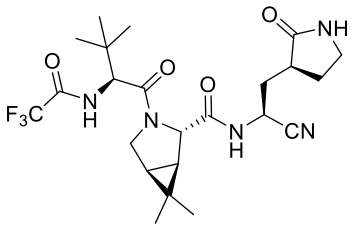
Abstract. A convergent route to the antiviral drug nirmatrelvir is described, arriving at the targeted drug in 45% overall yield with no loss of stereointegrity. Critical amide bond-forming steps utilize green technology that avoids traditional peptide coupling reagents. Likewise, dehydration of a primary amide to the corresponding nitrile is performed under environmentally benign conditions *in water* that avoids use of the Burgess reagent. Comparisons with the original literature procedures described highlight the decrease in environmental footprint associated with this approach.

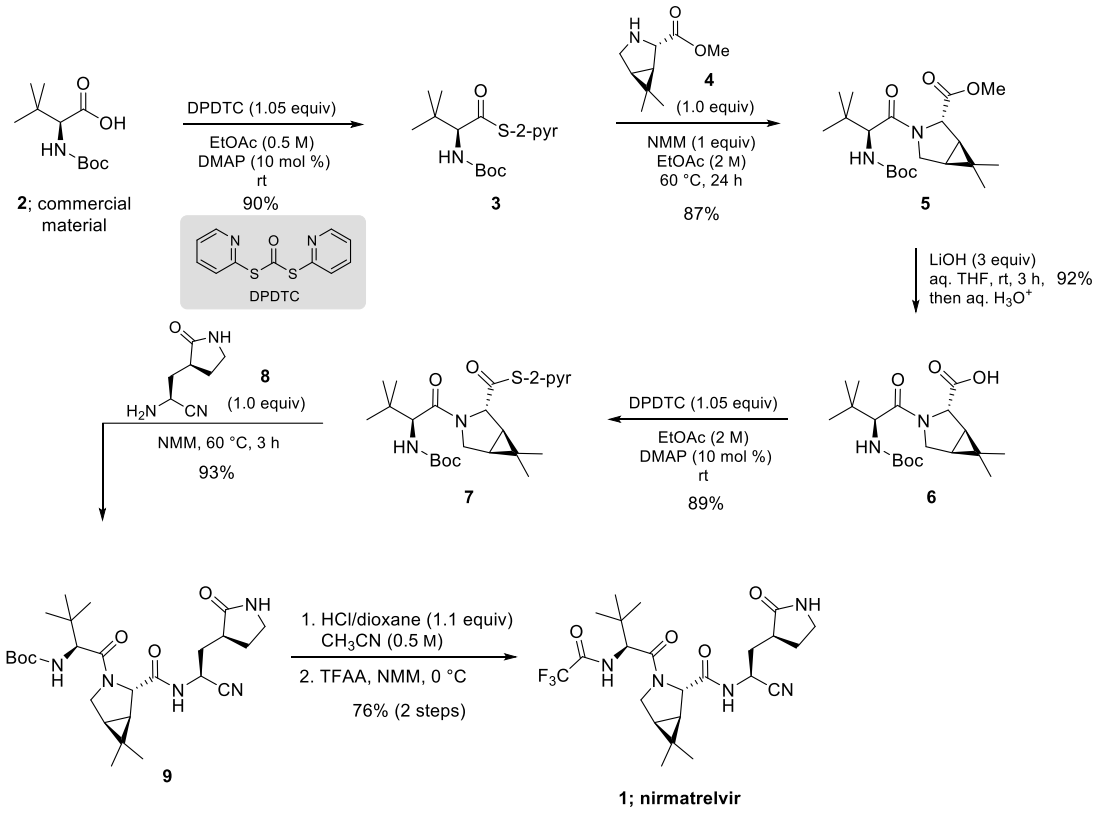
Introduction

The ongoing COVID-19 pandemic caused by the SARS-CoV-2 virus and its variants represents a once in a century public health disaster that continues to impact mankind, as well as the global economy.[1] The disease has created an urgent need for the rapid development of both preventative measures (i.e., vaccines) and post-infection treatment options.[2] Both aims have been achieved in record time, the latter initially being driven by the investigation into repurposing existing drugs, which has produced few effective treatment options.[3] The search for novel SARS-CoV-2 antivirals, on the other hand, has led to the development of molnupiravir by Merck,[4] EDP-235 by Enanta,[5] S-217622 by Shionogi,[6] and several others.[7] The first such antiviral to receive approval by the FDA, Paxlovid, is an orally-active combination of the SARS-CoV-2 main protease inhibitor nirmatrelvir (**1**; PF-07321332) and the HIV antiviral ritonavir, disclosed by Pfizer in late 2021.[8] This drug combination was shown to reduce risk of progression to severe COVID-19 in high-risk, symptomatic patients by 89% compared to placebo,[9] exemplifying the crucial role the drug is expected to play in the continuing efforts to combat the COVID-19 pandemic.

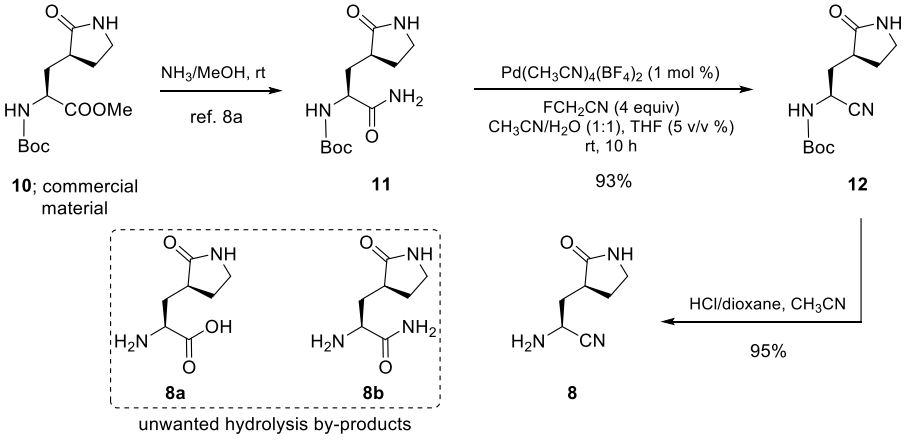


1; nirmatrelvir

The environmental impact of the synthesis of a drug in such immediate and high demand cannot be overlooked, especially since the prescribed dosage is 3 g of total API per patient over the course of treatment. While Pfizer's more recently reported route to nirmatrelvir[10] improves upon their originally reported methodology,[8a] there are still opportunities to reduce the amount waste generated by their use of organic solvents and peptide coupling reagents. Therefore, there exists an especially urgent need for the development of both a green as well as economically attractive synthesis of nirmatrelvir.



Scheme 1. The overall route to nirmatrelvir.



Scheme 2. Preparation of aminonitrile **8**.

Results and Discussion

In continuing our group efforts to develop scalable routes to APIs under environmentally friendly conditions,[11] and in an ongoing partnership with the Bill and Melinda Gates Foundation initially formed for purposes of preparing APIs for the treatment of malaria (e.g., pyronaridine),[11a] we have developed a route to nirmatrelvir that simultaneously addresses both of these issues (i.e., fiscal cost and environmental impact). Our approach is characterized by the development and application of green technologies, as well as minimization of both time and pot economies.[12] Furthermore, special attention has been directed towards eliminating the epimerization of chiral centers during crucial peptide bond-forming steps. Workups, which often give

rise to enormous volumes of organic waste, have also been streamlined to now involve only simple, in-pot aqueous washes, as well as use of minimal amounts of recoverable and environmentally preferred organic solvents (e.g., EtOAc).

The strategy selected for the synthesis of nirmatrelvir focused on the inherent benefits of convergent syntheses on scale.[13] Additionally, it was advantageous to perform our Pd-catalyzed amide dehydration in this convergent fashion, as subsequent steps provide opportunities to limit the amount of residual Pd in the final product.

The route begins with commercially available *N*-Boc-protected *t*-leucine (**2**) that is converted to its thioester derivative **3** using di-2-pyridyldithiocarbonate (DPDTC)[14] in environmentally preferrable EtOAc[15] containing catalytic DMAP at rt (90%; Scheme 1). Product **3** was purified via simple in-flask aqueous workup, followed by addition of bicyclic proline **4** and mild heating which led to the desired peptide bond in product **5** (87%). This 1-pot technology, which is being developed mainly for use under aqueous micellar conditions, avoids traditional peptide coupling reagents (e.g., HATU, DCC, COMU, T3P, etc.), eliminates epimerization, and in this case, allows for facile removal of the 2-mercaptopyridine by-product via an in-flask extraction with aqueous hydroxide. In contrast to the by-products of conventional amide bond coupling reagents, 2-mercaptopyridine can be easily recovered for recycling to DPDTC (see ESI Section 4). The newly formed peptide as its methyl ester is then hydrolyzed with LiOH in aqueous THF, after which the reaction mixture is neutralized with aqueous HCl and then extracted with minimal EtOAc to give **6** (92%). The resulting acid **6** is then subjected to thioester/peptide bond construction (*vide supra*) en route to **9** (93%) via **7** (89%). For this peptide coupling between **7** and **8**, the mild base *N*-methylmorpholine (NMM) was used to neutralize the HCl salt of **8**. It should be noted that both intermediate thioesters **3** and **7** are stable, isolable species, although in this sequence their individual isolation/purification was not needed.

The aminonitrile **8** to be used in the second peptide-forming step was prepared from commercially available material: the *N*-Boc-protected methyl ester **10**, which was smoothly converted to the corresponding primary amide **11** using the published procedure (methanolic ammonia; Scheme 2).[8a] Although previous reports for the dehydrative conversion of **11** to the required nitrile (**12**) utilized the Burgess reagent in chlorinated solvent, primary amide **11** could be smoothly dehydrated in water applying recently disclosed technology based on "amide exchange."[16] That is, where commercially available fluoroacetonitrile served as the sacrificial acceptor of water under Pd-catalyzed conditions. The newly fashioned nitrile **12** (93%) was then treated with HCl/dioxane in CH_3CN to effect Boc removal leading to the amine salt of **8** for use in the second peptide coupling, as described above.

It is important to note that Boc-deprotection of **12** using HCl in organic solvents was highly variable in that adventitious water present both in the solvent and in starting material **12** led to varying amounts of hydrolysis to form carboxylic acid **8a** and/or primary amide **8b** (13-30+%). It was anticipated that hydrolysis would also occur during the Boc-deprotection of **9** prior to conversion of the free amine with TFAA to nirmatrelvir (**1**), and thus a method which gives rise to minimal hydrolysis was required. Formation of by-products **8a** and **8b** could be minimized via azeotropic removal of residual water in **12** using toluene under high vacuum. Prior observations by BMS on related nitriles indicated that inclusion of a sacrificial nitrile, such as CH_3CN, reduced undesired competitive hydrolysis during Boc-deprotection, likewise under acidic conditions.[17] Indeed, applying both procedures (azeotropic drying of the educt and then adding dry CH_3CN) afforded the desired nitrile amine as its HCl salt (95%) with only 3% hydrolysis. Separation and removal of residual **8a** and/or **8b** could be easily accomplished by dissolving the mixture in MeOH and precipitating pure **8** using ice-cold Et_2O.

Similar Boc-deprotection conditions could then be applied to intermediate **9** to afford the amine HCl salt. Following removal of solvent and excess HCl *in vacuo* and resuspension in CH_3CN, the material was subsequently exposed to TFAA containing NMM as base. Removal of excess TFAA and NMM via aqueous washes, followed by column purification, afforded nirmatrelvir (**1**; 76% over 2 steps).

Overall, this convergent sequence afforded nirmatrelvir in 45% overall yield, comparable to the 48% overall yield disclosed by Pfizer.[8a] Nonetheless, Table 1 summarizes a direct comparison of several additional key features associated with each route with respect to environmental considerations, and in all likelihood, costs. Efforts are currently underway to further optimize the individual steps, as well as to perform this sequence in as few as 2-pots over 7-steps, which will further reduce the environmental footprint of the process, especially on scale where a separate pot for each step necessitates use of copious volumes of organic solvents for reactor cleaning. This improved multistep 2-pot approach will be disclosed in due course.

Table 1: Comparisons with Pfizer's route[8a]

synthetic step	Pfizer	this work
amide bond formations	• HATU, EDCI • non-recyclable waste • solvents: DMF, MEK	• **DPDTC** • **recyclable 2-mercaptopyridine** • **solvent: EtOAc**
amide dehydration	• Burgess reagent • solvent: CH_2Cl_2	• **cat. Pd, FCH_2CN** • **solvent: H_2O/CH_3CN**
Boc-deprotection	• solvent: CH_2Cl_2	• **solvent: CH_3CN, MeOH**
overall yield	48%	45%

Summary

The sequence to nirmatrelvir outlined herein provides streamlined, efficient, convergent, and environmentally responsible access to a highly valued drug for treatment of COVID-19. The route features:

• peptide bond constructions that take place in a 1-pot process in highly concentrated EtOAc that avoid traditional peptide coupling reagents which can be costly, dangerous, and produce considerable waste, especially at scale

• application of a newly developed, green technology for amide dehydration applied to a "real" molecule, that by-passes use of unattractive reagents (e.g., Burgess reagent)

• conditions that avoid potentially costly separation of unwanted isomers due to epimerization

• a greatly reduced environmental footprint, thereby avoiding much of the waste being generated by currently utilized routes

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Notes

The authors declare no competing financial interest.

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