Design and Synthesis of antiviral Molnupiravir analogues

Qi Chen, Jarod M. Marasco, Jenna N. Kriley*
Department of Chemistry, Slippery Rock University, Slippery Rock, PA 16057

Introduction:

Viral infectious diseases have been proved to be a major threat to our public health, including the ongoing COVID-19 pandemic. Two pillars of fighting viral infectious diseases are vaccines and antiviral drugs. Despite the immense achievements of our COVID-19 vaccines, yet the pace of antiviral drug development is dreadfully slow. Besides Remdesivir, only two more compounds (currently under development by Pfizer and Merk respectively), are showing significant progress in treating COVID infections. Two of three (Remdesivir from Gilead and Molnupiravir from Merck) (Figure 1) are nucleosides analogues, which indicate the importance of nucleosides analogues in the field of antiviral research. Their antiviral mechanism could be rather varied despite the chemical structure similarity among the nucleoside analogs. Such as Redemisvir stops the viral replication by halting the RNA-dependent RNA polymerase (an RNA assembling machine). In contrast, Molnupiravir, an RNA mutagen, causes the virus to accumulate replicating errors, thus leading to viral death. Sometimes, structure differences as minor as changing one atom could respond to the diverse antiviral mechanism. And diversity is the key to battling viral resistance to the exiting antiviral drugs.

Figure 1 The structures of Molnupiravir and Remdesivir

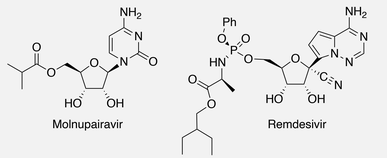
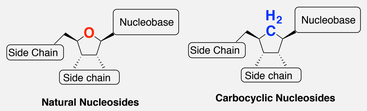


Figure 2. Naturally Occurred and Carbocyclic Nucleosides



The presented study is the first in the line to pursue a series of chemically modified compounds designed based on the antiviral drug Molnupiravir (EIDD-2801, MK-4482), currently finished phase II/III clinical trials with a promising positive result and was granted an emergency use authorization by U.S. FDA. The newly designed targets adopted the carbocyclic sugar framework to improve the antiviral activity by increasing the cyto-stability compared to the parent compound. Carbocyclic nucleosides (Figure 2) are also known for their prominent board-spectrum antiviral activities with distinct drug action mechanisms. Their antiviral activity stems from inhibition of a host enzyme Adenosylhomocysteine (AdoHcy) hydrolase, then consequentially stop the "capping" progress, a major step in forming the mature viral mRNA (Figure 3). The designed compounds (Figure 4) combine the features of Molnupiravir (nucleobase) with the carbocyclic rings to pursue a class of dual antiviral mechanism drug candidates. Potential pro-drug structures are also included to test the pharma kinetic properties. The proposed synthesis strategy successfully achieves the key intermediate via a Mitsunobu coupling reaction. Optimization of the reaction conditions for the critical synthesis steps is detailed in the following discussion.

Figure 3. Antiviral Mechanism of AdoHcy Hydrolase inhibitors

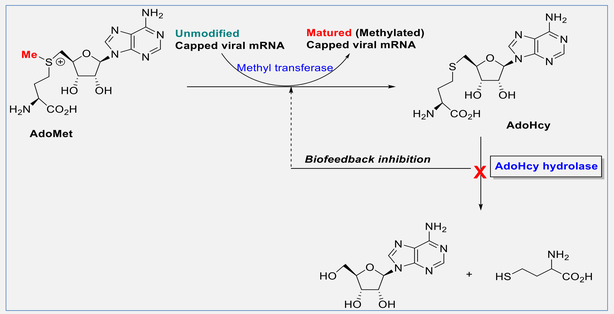
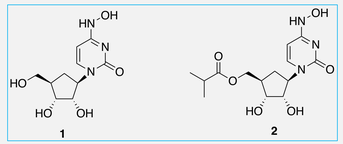
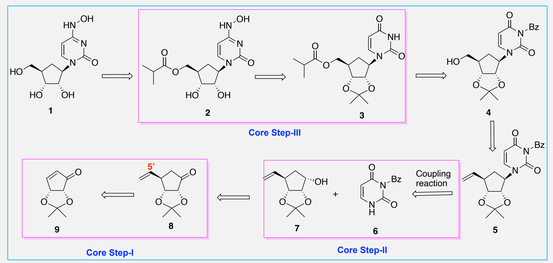


Figure 3. Designed Target Compounds, Carbocyclic Molnupiravir

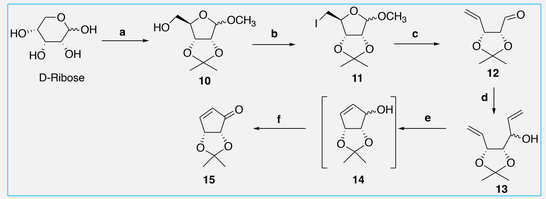


Chemistry:

The designed synthesis of target compounds **1** and **2** includes three major synthetic challenges we presented here as three core steps in synthesis (Scheme 1). The first challenge (core step I) involves establishing the 5'-side chain via a 1,4-Michael addition of a vinyl group to the key intermediate enone **9**, obtained through a 6-step synthesis by the undergraduate researcher (Scheme 2).

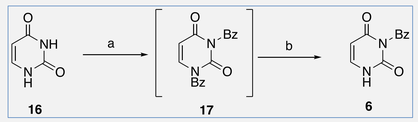


Scheme 1: Retrosynthesis design for target compounds



Scheme 2. Synthesis of intermediate **15**: a.$(MeO)_2CMe_2$, MeOH, HCl, acetone, rt., 72%; b. PPh_3, I_2, imidazole, 86%; c. Zn, MeOH, 96%; d. vinylmagnesium bromide, THF, 78%; e. Grubbs catalyst, CH_2Cl_2; f. PDC, CH_2Cl_2, 90%.

The second challenge (core step II) is to combine nucleobase **6** with pseudo sugar **7** via a coupling reaction. Two main components in this step (**6** and **7**) are synthesized parallelly to improve success rate. The strategy is known as convergent synthesis in comparison with linear synthesis. The synthesis of nucleobase coupling precursor **7** is shown below in scheme 3.



Scheme 3. Synthesis condition for compound **6**: a. Benzoyl chloride, acetonitrile/pyridine (5:2, v/v); b. K_2CO_3, dioxan/H_2O (1:1, v/v), CH_2Cl_2, 77% in two steps.

The first attempt to construct the 5'-side chain is to use the in-suit formed Gilman reagent under the low temperature to facilitate the 1,4-addition of unsaturated ketone **9**. No reaction under -75°C, even after stirring overnight. After rising the temperature to -20°C, compound **8** was formed, accompanied by the 1,2-addition by-product **8b** (Scheme 4). The other condition (cond. 2) was also tried using less reactive vinyltrifluoroborate as a nucleophile with rhodium catalyst. Compound **8** was achieved with a low yield (27%). After reduction (b) and Mitsunbu coupling (c) compound **5** was obtained as a low quantity mixture and difficult to carry on for the later synthesis.

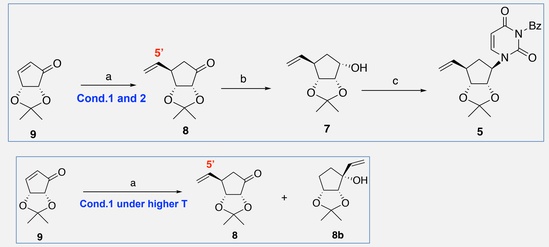
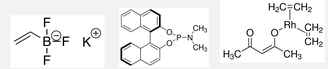


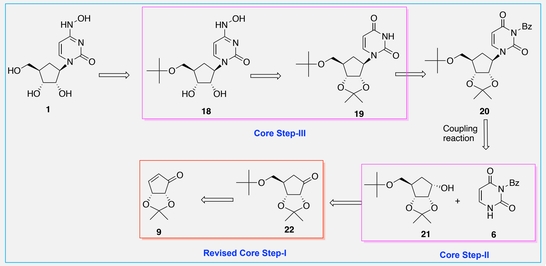
Figure **4**:Structures for the reagents in a. condition 2



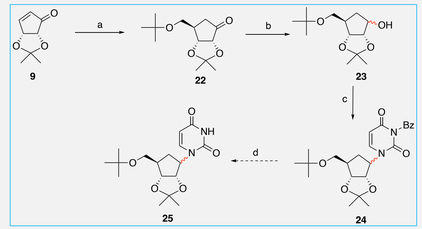
Scheme 4. Synthesis condition for compound **5**: a. condition 1: vinylmagnesium bromide,CuBrMe_2S, TMSCl, HMPA, THF, -78°C; condition 2: acetylacetonatobis(ethylene)rhodium (I), (*R*)-MonoPhos, potassium vinyltrifluoroborate, EtOH, reflux; b. $LiAlH_4$, THF, 0°C; c. compound **6**, DIAD, PPh_3, 60°C.

The new synthesis strategy (Scheme 5) is pursued to optimize the 5' side chain construction (Core step-I). The protected alkoxyl group is chosen as the nucleophile, which will shorten the synthesis route by two steps (oxidative cleavage and reduction) compared with the old design (Scheme 1).

The 1,4-addition of the 5' side-chain was successful installed by a in situ formed Gilman reagent transformed from a lithium reagent. The ^{1}HNMR below shows the achieved pure compound **22**. The reduction using the $LiAlH_4$ gave a mixture of diastereomers of the hydroxyl group on the 1' position. The condition will be modified to use less reactive $NaBH_4$ to ensure the enantiomeric pure compound **23**.



Scheme 5. Revised Retrosynthesis design for the target compound **1**.



Scheme 4. Synthesis condition for compound **22**: a) *t*BuOK, *t*BuOMe, *sec*-BuLi, $CuBrSMe_2$, diisopropyl sulfide, THF. B) LiAlH4, THF 0°C.

Acknowledgements:

We are grateful to the FSRG Research Grant fund from Slippery Rock University for their support of this research.

References:

1. (a) Patil, S. D.; Schneller, S. W.; Hosoya, M.; Smpeck, R.; Andrei, G.; Bbalzarini, J.; De Clercq, E. *J. Med. Chem.* **1992**, 35, 3372; (b) Siddiqi, S. M.; Chen, X.; Schneller, S. W.; Ikeda, S.; Snoeck, R.; Andrei, G.; Balzarini, J.; De Clercq, E. *J. Med. Chem*. **1994**, 37, 551; (c) Yang, M.; Schneller, S. W. *Bioorg. Med. Chem. Lett*. **2005**, 15, 149; (d) Yang, M.; Schneller, S. W.; Korba, B. *J. Med. Chem*. **2005**, 48, 5043.
2. Schneller S. W. *Rec Adv. Nucleotides*. **2005**; 24:1395.
3. Liu C.; Chen Q.; Schneller S. W. *Bioorg. Med. Chem. Lett.* **2016**, 26, 928.
4. Schneller S. W.; Liu C.; Chen Q.; Ye. W. PCT Int. Appl. 2016, WO 2016022563 A1; US 9,657,048.
5. Chen Q.; Davidson A. *Bioorg. Med. Chem. Lett.* **2017**, 4436.
6. (a) De Clercq, E. *Nucleotides Nucleic Acids* **2005**, 24, 1395. (b) Liu, C.; Chen, Q.; Schneller, S. W. *Bioorg. Med. Chem. Lett.* **2012**, 22, 5182.
7. (a) DeClercq, E. *J. Clin. Virol*. **2004**, 30, 115. (b) Agrofoglio, L.; Suhas, E.; Farese, A.; Condom, R.; Challand, S. R.; Earl, R. A.; Guedj, R. *Tetrahedron* **1994**, 50, 10611. (c) Huryn, D. M.; Okabe, M. *Chem. Rev*. **1992**, 92, 1745.
8. Liu C.; Chen Q.; Yang M.; Schneller S. W. *Bioorg. Med. Chem*. **2013**, 21, 359.
9. Liu C.; Chen Q.; Schneller S. W. *Bioorg. Med. Chem. Lett.* **2012**, 22, 5182.