A Concise Biocatalytic Synthesis of Pretomanid.

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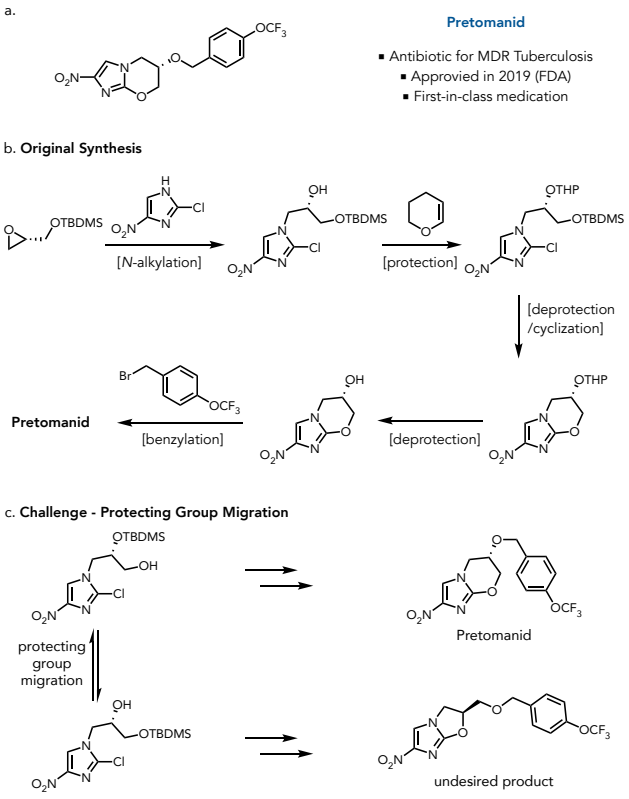
ABSTRACT: An efficient protocol to produce Pretomanid is proposed and studied. The stereodetermining step of the proposed synthesis a ketone reduction by a ketoreductase. However, the synthesis of the proposed ketone proved challenging because of the unexpected stability of the ketal intermediate.

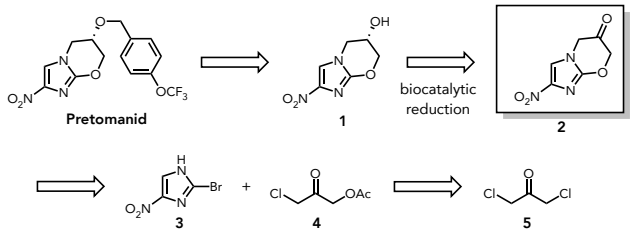
Tuberculosis (TB) is an ancient disease that kills more than 1.8 million people each year worldwide.[1] It is caused by *Mycobacterium tuberculosis* (Mtb) and the emergence of multidrug resistant and extremely drug resistant strains has made this pathogen a persistent problem. Pretomanid was originally identified in a series of nitroimidazopyran derivatives tested for antitubercular activity (Scheme 1a).[2] In 2019, it was approved by the FDA for the treatment of a limited and specific population of adult patients with extensively drug resistant, treatment-intolerant or nonresponsive multidrug resistant pulmonary tuberculosis (TB). The development of a more efficient and cost-effective synthesis of Pretomanid is eagerly needed as it would enable the broader distribution of this medicine.

Scheme 1. Original Synthesis Route of Pretomanid.



The original synthesis of Pretomanid employs a chiral epoxide precursor and involves several protecting group manipulations (Scheme 1b).[3] However, undesired side product that is difficult to separate from the targeted product would be generated during this route and complicates the isolation efforts. This is due to the migration of the TBDMS protecting group under basic reaction conditions (Scheme 1c). Therefore, we aimed to develop a protecting-group free route to Pretomanid, which would be valuable and greatly increases the overall efficiency of the synthesis. Our designed pathway employs commercially available 1,3-dichloropropan-2-one (**5**). Conversion to the mono-acetoxy ketone followed by imidazole alkylation and cyclization would afford a cyclic ketone **2**. We proposed that a biocatalytic asymmetric ketone reduction by a ketoreductase (KRED) to set the molecules sole stereocenter. KREDs are idea for this type of reduction because they can afford high efficiency, selectivity and evolvability of KRED.[4]

Scheme 2. Proposed Synthesis of Pretomanid.

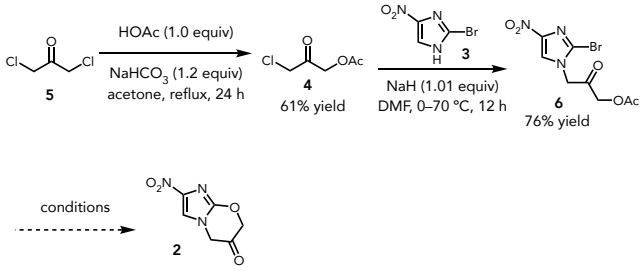


We began by looking at the synthesis of **2**. Treating 1,3-dichloropropan-2-one (**5**) with acetic acid in the presence of sodium bicarbonate in reflux acetone afforded the 3-chloro-2-oxopropyl acetate (**4**) in good yield (Scheme 3).[6] Then the imidazole derivative (**3**) was deprotonated and attacked **4** to generate **6** efficiently. However, all attempts to access the ketone compound **2** via deacylation and cyclization were unsuccessful, presumably due to the fast decomposition of **6** to imidazole starting material under basic conditions. To solve this problem, we designed an alternative method to access **2**.

We envisioned that if the carbonyl group was protected before treating with base, the deacylation and cyclization would proceed smoothly, and **2** would be generated after deprotection. We employed a two-step one pot protocol. **6** was reacted with trimethyl orthoformate in the presence of catalytic amount of acid first, then K_2CO_3 was added and the reaction was stirred at rt overnight (Scheme 4). The corresponding ketal **7** was formed

in good yield. To our surprise, the deprotection of **7** to yield **2** turned out to be quite challenging. A number of Bronsted acids, Lewis acids were investigated and all failed to afford the desired product (Table 1, entry 1-6).[7] Oxidative condition using DDQ was also attempted but was not successful neither (Table 1, entry 7). We were intrigued by this observation and performed cryptoscopic analysis on **7** to confirm its structure. We then studied the reactivity of compound **8** and **9** (Scheme 5), and found them to be extremely stable under ketal deprotection conditions. Currently we still were not able to fully understand this phenomenon but the unique geometry of the fused six-membered ring might play an important role in stabilizing these ketal complexes.

Scheme 3. First Attempted Synthesis of 2.



Scheme 4. Alternative Synthesis of 2.

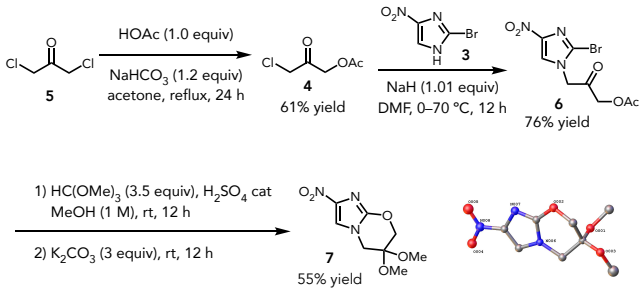
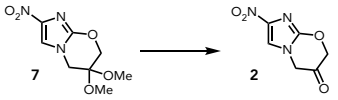


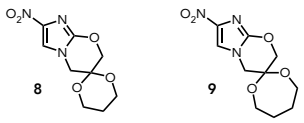
Table 1. Representative Deprotection Conditions Tested.



entry[a]	reagent	temp	results[b]
1	TsOH (2 equiv.), acetone (0.5 M)	rt	no reaction
2	AcOH (2 equiv.), THF:H_2O = 1:1 (0.2 M)	45	no reaction
3	TFA (0.2 M)	rt	no reaction
4	I_2 (2 equiv.), acetone (0.5 M)	rt	no reaction
5	TMSI (2 equiv.), DCM (0.5 M)	rt	**7** fully decomposed, no product
6	BBr_3 (2 equiv), DCM (0.5 M)	rt	no reaction
7	DDQ (2 equiv.), MeCN:H_2O = 1:1 (0.5 M)	rt	no reaction

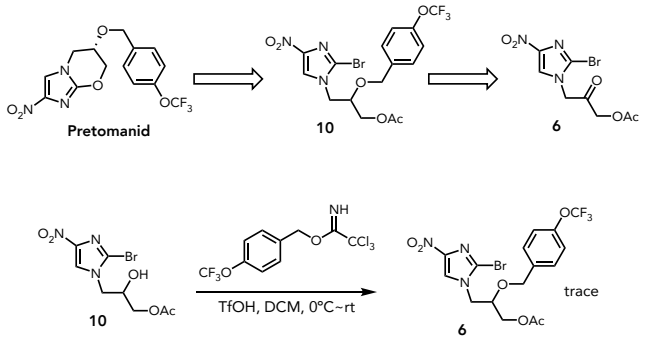
a, All the reactions were performed at 10 mg (**7**) scale. *b*, no reaction means that the conversion of **7** is < 95%, no product is formed.

Scheme 5. Other Protecting Group Investigated.



Realizing the difficulty in accessing the ketone **2**, we proposed an alternative route for the synthesis of Pretomanid. We envisioned that from **6**, if the carbonyl was reduced and benzylated first, the formation of **2** would be obviated and Pretomanid would be afforded after deacylation and cyclization (Scheme 6). In the model reaction using racemic alcohol **10**, we found that the benzylation had to be performed under acidic conditions to avoid the migration of the acetyl group. However, the imidazole ring was not compatible with acidic benzylation conditions and the desired product was only formed in trace amount.

Scheme 6. Alternative Route of Pretomanid.



In summary, an improved synthesis of Pretomanid, involving a novel ketoreductase-catalyzed asymmetric reduction was described and studied. The biocatalytic step was investigated and proved feasible. However, an unusual stabilizing effect observed during the deprotection of a ketal complex prevented the formation of the key product precursor and resulted in an unsuccessful synthesis. Studies on unique reactivity of the ketal compound is undergoing and alternative routes to Pretomanid are under investigation.

ASSOCIATED CONTENT

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Notes

The authors declare no competing financial interest.

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