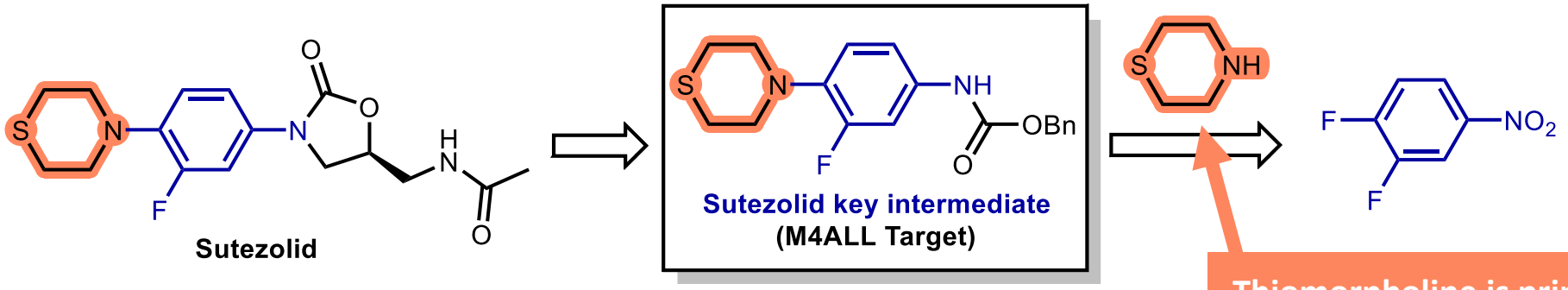
Improved Process to Prepare a Key Intermediate in the Synthesis of TB Drug Sutezolid

September 2023

Background and Executive Summary

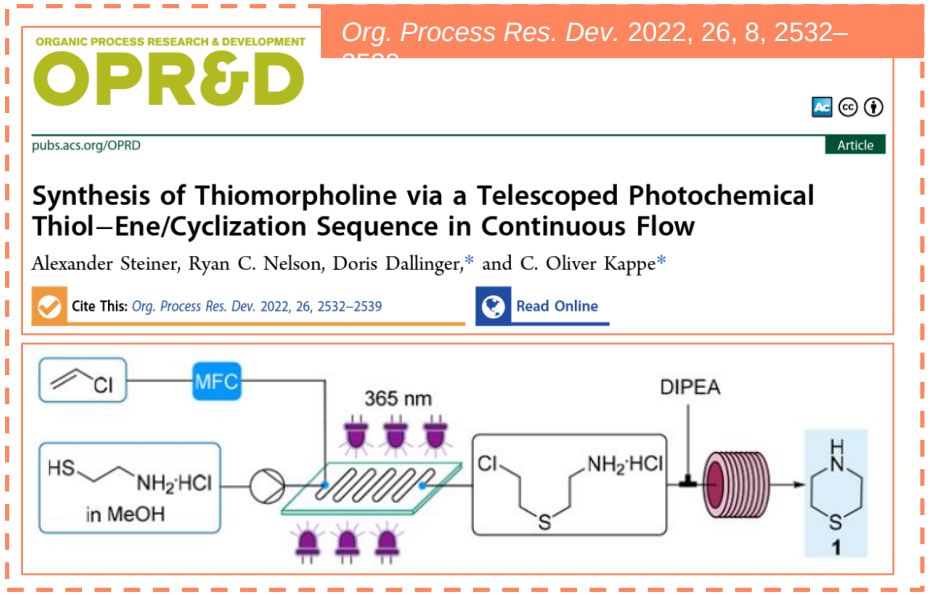
Defining the Problem with Sutezolid Synthetic Process



Thiomorpholine is primary cost driver because of low production volumes and hazards present in its manufacturing process

Scope of M4ALL Work:

1) Investigate safer processes to make **thiomorpholine** (collaboration with Kappe Group, Univ Gratz);
2) Develop a process to make key **Sutezolid key intermediate** by avoiding thiomorpholine and leveraging existing commodity chemicals (collaboration on scaleup with TCG Life Sciences).

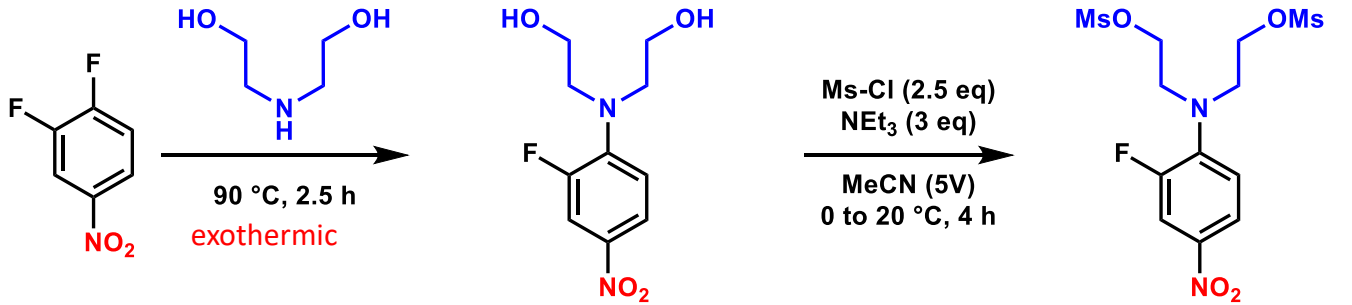


M4ALL Process Summary

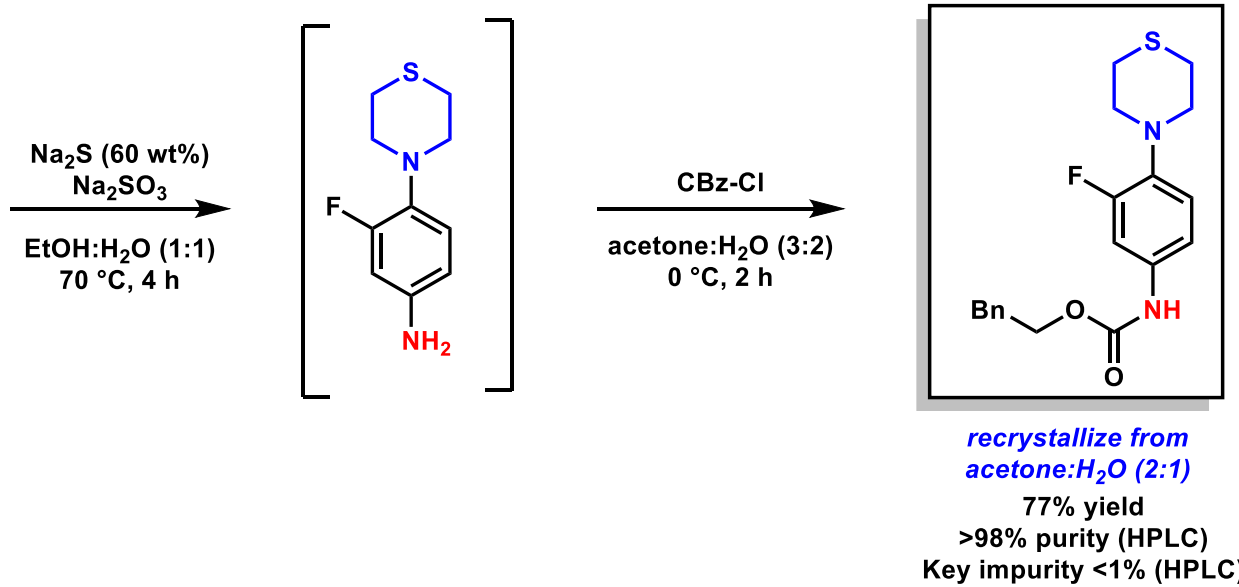
Key Developments:

- Utilization of commodity diethanolamine as S_NAr partner, thus avoiding expensive thiomorpholine
- Elimination of the Pd/C-catalyzed reduction, which was the second largest cost driver
- Scalable end-to-end synthesis of Sutezolid key intermediate on >100 g scale (successfully reproduced by TCG Life Sciences)
- Development of a purification strategy that delivers key intermediate in ≥98% purity and >55% overall yield
    - The most impactful impurity, a morpholino derivative, was reduced to <1% (HPLC)

Step 1 ***Step 2***



Telescoped Steps 3/4



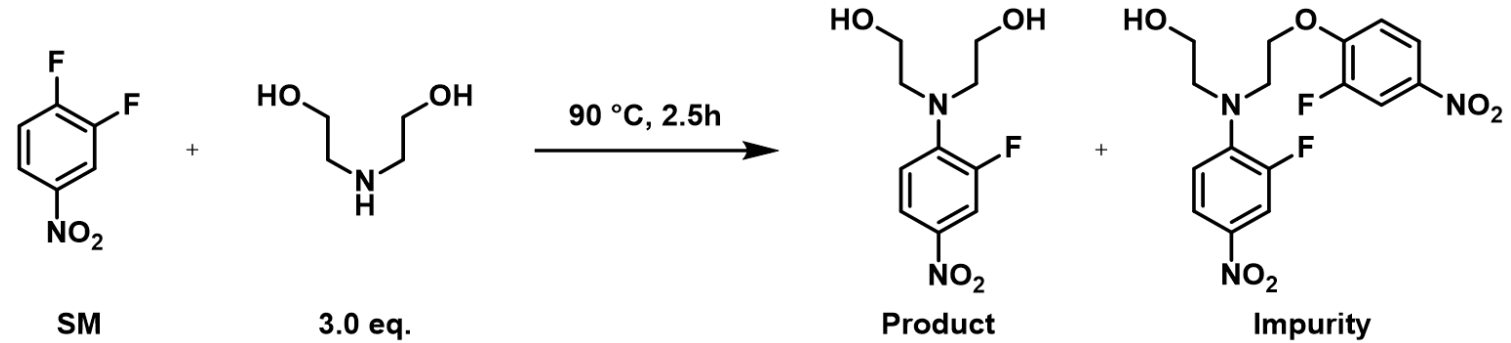
DEFINING IMPACT

Potential to reduce raw material cost by up to ~80% to this valuable intermediate versus the baseline thiomorpholine route*

**based on thiomorpholine costs represented in bills of lading (import/export in China & India)*

M4ALL Synthetic Process

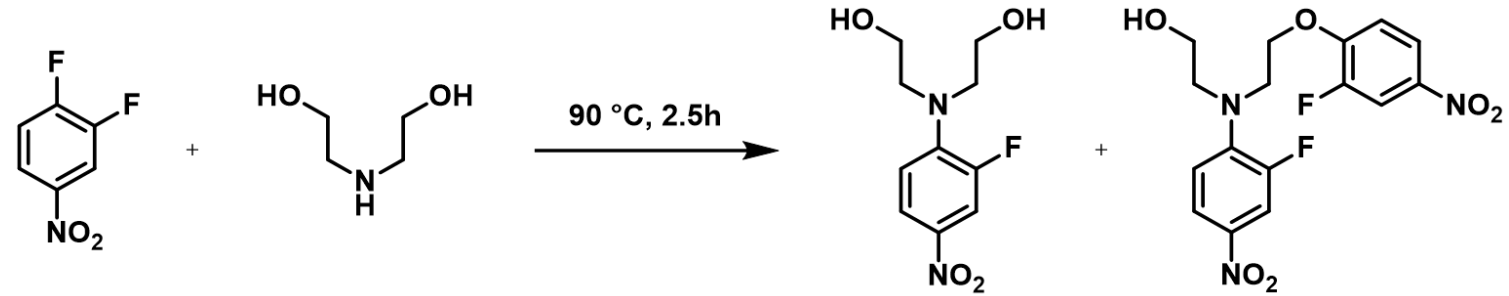
Step 1: Current Best Procedure for Crude Product



SM 3.0 eq. Product Impurity

Batch scale	Procedure	Analytical data	Isolated Material
500.0 g	Diethanolamine (991.3 g, 3.0 eq, corresponds to ~900 mL or ~1.8V) was heated to 90 °C (int. temp.). After maintaining constant temp. for 5 minutes, difluoro SM (500.0 g, 1 eq) was added to the reaction mixture with a dropping funnel over 1.5 h. *SAFETY NOTE:* This addition is exothermic, so reaction temperature should be monitored closely with reagent addition. The reaction mixture was maintained at temperature for another 2.5 h after addition. Reaction completion was determined by IPC. Isolation: The heating was stopped, and the reaction mass was allowed to cool to 25-30 °C (~3.5 h). Water (5V, 2500 mL) was added to reaction mixture in 4 lots. (Solid formation was observed after 2nd addition.) The reaction slurry was stirred for 2 h and then cooled to <5 °C. After 4 h at this temp, the yellow solid product was filtered through Buchner funnel, washed with H_2O (1000 mL, 2V). Suck dry the product for 2 h before taking the product to the purification stage.	**IPC (2.5 h)** **LCAP** Product = 90.6 Impurity = 4.71 SM = ND IPC	**Crude Wt.:** 1005 g Product LCAP = 92.3 Impurity LCAP = 5.81 KF = 21.83% (w/w) Crude Crude KF

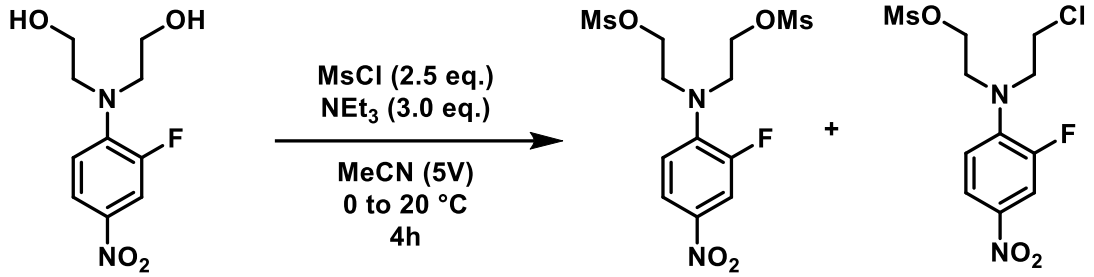
Step 1: Current Best Procedure for Purification



SM | 3.0 eq. | Product | Impurity

Batch scale	Procedure	Isolated Material (First Crop)
1005 g (Crude rxn product)	**First crop:** The crude product (1005 g, 21.83% water) was transferred to a RBF and charged with EtOAc (3.5V, 1750 mL based on original batch size). This was heated to reflux (75-80 °C), at which point the solution should clarify. This temperature was maintained for 2 h before cooling back to RT, at which point solid formation should begin. Further cooled the mixture to 0 – 5 °C and maintained for 3 h. Filter the yellow solid and wash with chilled EtOAc (1V at 5 – 10 °C, 500 mL based on batch size). Continued to suck the product dry for 1 h before drying under vacuum at 50 – 55 °C for 8 h. **Second crop:** A second crop of material could be isolated by distilling off the EtOAc from mother liquor and repeating the crystallization process. The amount of EtOAc should be scaled to the amount of solid recovered from the mother liquor. The quality of this crop (67.5 g) is slightly lower (Product LCAP = 96.26) than the first.	**Crude Wt.:** 675 g **Yield:** 85.9% (assay corrected) **LCAP** Product = 99.83 Impurity = 0.15 SM = ND Pure KF = 0.28% (w/w) Assay = 97.72% by qNMR QNMR Pure-NMR Pure-KF Pure-DSC

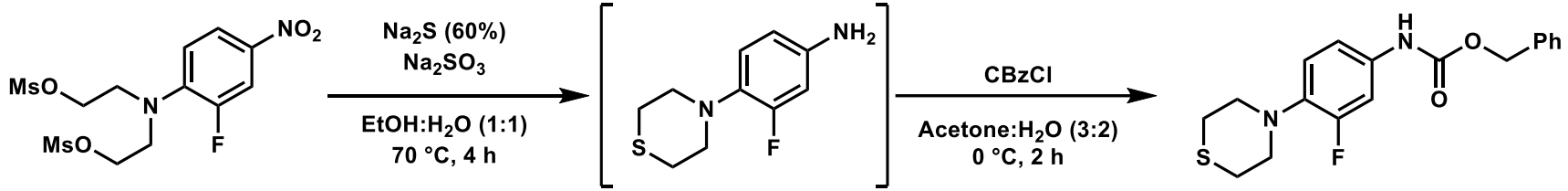
Step 2: Current Best Procedure



Impurity

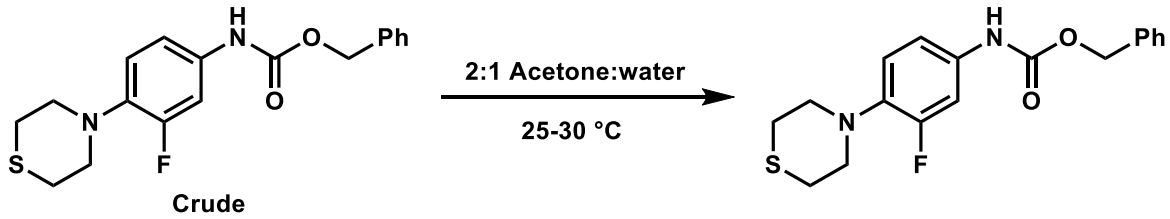
Batch scale	Procedure	Analytical data	Isolated Material
500 g (99.83% purity, previous slide)	A flask containing the diol derivative (500.0 g, 1.0 eq.) under a N_2 atmosphere was charged with acetonitrile (2500 mL, 5V) and triethylamine (621.6 mL, 3.0 eq.) at RT. The stirred reaction mixture was cooled to an internal temperature 0-5 °C. Methane sulfonyl chloride (586.3 g, 2.5 eq.) was added dropwise over 2 h while an internal temperature below 15 °C was maintained. The reaction mixture was allowed to warm up to RT and stirred for 4 hours at that same temperature. At this point, a sample of the reaction mixture was submitted for IPC. Isolation: Water (2000 mL, 4V) was added at once to the reaction mixture, and the reaction mass (slurry) was stirred for overnight (16 hours) at room temperature (20-25 °C). The reaction mass was cooled to 0-5 °C using an ice bath and stirred for additional 2 hours. The solids were isolated by vacuum filtration and washed with ice-cold water (1000 mL X 2) followed by MTBE (1000 mL X 2). The solids were then dried under vacuum for 6 h at 50 °C to afford a yellow solid.	**IPC LCAP** Product= 85.97% IPC	**Crude Wt.:** 689.5 g **Yield:** 86.35% (Assay corrected) **LCAP** Product = 97.82 SMI = 1.06 Crude Crude NMR assay ROI

Steps 3/4: 100 g Telescoped Batch



Batch scale	Procedure	IPC Data	Isolated Material
100 g (98.32 LCAP, 97.7% Assay)	A 5 L RBF with a mechanical stirrer (185 RPM), N_2 inlet, and condenser was charged with a solution of dimesyl derivative (100.0 g, 1.0 eq.) dissolved in EtOH:water (1:1 ratio, 500 mL, 5V). This mixture was heated to an internal temp of 75-80 °C with stirring, at which point Na_2SO_3 (15.7 g, 0.5 eq) was added. This was followed by dropwise addition, over 15 minutes, of a solution of Na_2S (60%, 97.5 g, 3.0 eq) dissolved EtOH:water (1:1 ratio, 1000 mL, 10V). Stir this mixture at temperature for an additional 4 h. After completion of the reaction (IPC 1: SM LCAP below detection), the reaction mixture was cooled to 25-30 °C, and the pH was adjusted to 10-11 with 1 N HCl (~5 V). EtOH was distilled under vacuum from the reaction mass (~8.5 V reaction mass remining. Acetone (10 V, 1000 mL) was added, and the reaction mixture was cooled to 0-5 °C. CBzCl (51.09 g, 1.2 eq), as a solution in toluene (25 mL, 0.25 V), was added dropwise over 60 min, and then the reaction was maintained at 10-15 °C for 2 h. After completion of the reaction (IPC 2: aniline LCAP below detection limit), water (10 V, 1000 mL) and heptane (2 V, 200 mL) were added, and the reaction was stirred for 4 h at 25-30 °C. Isolation: Cool the reaction mixture to 10-15 °C and stir for 2 h. Filter the precipitated compound. Wash the filtrate with water (10 V, 1000 mL) and heptane (2 V, 200 mL). Unload the product and vacuum dry at 40-45 °C.	**IPC 1 LCAP:** **SM =** BDL **Aniline** = 77.39 IPC-1 **IPC 2 LCAP:** **Aniline =** BDL **Product** = 51.93 **Morpholine** = 5.46 IPC-2	**Crude Wt.:** 82 g **Assay:** 76.5% **Yield:** 70.8% Assay corrected **LCAP:** **Product** = 91.62 **Morpholine** = 3.75 Crude-HPLC Crude-Assay

Step 4 Product Purification: 2:1 Acetone:Water



Crude

Batch scale	Procedure	Isolated Material
80 g Crude LCAP: 91.62 product; 3.75 morpholine. Assay: 76.5%	Crude compound (80 g) and acetone (800 mL, 10 V) at 25-30 °C were charged into a RBF with a magnetic stir bar. Water (400 mL, 5 V) was slowly added, at which point a solid begins to precipitate. This mixture was maintained at temperature with stirring for 3 h. Cool the system to 10-15 °C and maintain for an additional 1 h. The precipitated compound was filtered and washed with chilled water (400 mL, 5 V) followed by methanol (160 mL, 2 V) and n-heptane (160 mL, 2 V). The solid was then dried under vacuum at 45-50 °C until constant weight (~4 h).	**Crude Wt.:** 61.0 g **Assay:** 87%* **Yield:** ***61.4%*** Assay corrected (2 steps) **KF:** 4.64% **LCAP:** **Product** = ***97.70%*** **Morpholine** = ***0.82%*** CR592-17066-60-P1 CR592-17066-60-P1-Assay CR592-17066-60-P1 kf

- The assay for the purified compound was low presumably because of inorganic impurities and water
  - Full solubility of the crude material was *not* observed after addition of acetone
  - This was initially attributed to poor product solubility, but this result suggested undissolved inorganics
- In subsequent 300 g runs, the isolated crude product was not full dried under vacuum before purification, and the mixture obtained after addition of acetone (purification step) was clarified by filtration through celite
  - The celite was rinsed with additional acetone (2 V) prior to water addition (6 V, increased to retain the 2:1 ratio)
- Future improvements may be possible by optimization of acetone usage before the celite filtration

Steps 3/4: 300 g Telescoped Reaction Summary

Input	IPC Data	Crude Isolated	After Single Crystallization
300 g (98.94 LCAP, 96.2% Assay)	**IPC 1 LCAP:** **SM =** BDL **Aniline** = 81.84 **Morpholine** = 2.42 IPC **IPC 2 LCAP:** **Aniline** = BDL **Aniline** = 74.43 **Morpholine** = 3.83 IPC-2	**Crude Wt.:** 272 g **Assay:** 67.7 wt% **Yield:** 70.96% Assay Corrected **KF:** 17.87% Crude-HPLC **LCAP:** **Product** = 90.65 **Morpholine** = 1.42 CR592-17087-35-CR-assay CR592-17087-35-CR kf	**Weight:** 161.0 g **Assay:** ***98.2 wt%*** **Yield:** ***63.7%*** Assay corrected (~80% over two steps) **KF:** 0.4 wt% HPLC Assay **LCAP:** **Product** = ***97.93*** **Morpholine** = ***0.46*** LCMS CR592-17087-35-P1-KF
Mixture: 250 g (99.23 LCAP, 98.9% Assay) *plus* 50 g (98.32 LCAP, 97.7% Assay)	**IPC 1 LCAP:** **SM** = BDL **Aniline** = 86.3 **Morpholine** = 2.15 IPC-1 **IPC 2 LCAP:** **Aniline** = BDL **Aniline** = 75 **Morpholine** = 7.55* IPC-2 * The blank for this run showed a peak at the same RT, so this value may be too high.	**Crude Wt.:** 284 g **Assay:** --- wt% **Yield:** ---% Assay Corrected **KF:** 19% **LCAP:** **Product** = 95.29 **Morpholine** = 2.29 crude HPLC	**Weight:** 174 g **Assay:** --- wt% **Yield:** ---% Assay corrected (over two steps) **KF:** 0.21 wt% HPLC **LCAP:** **Product** = ***98.51*** **Morpholine** = ***0.57***