ORGANIC PROCESS RESEARCH & DEVELOPMENT
OPR&D

Full Paper

Scale up synthesis of IID572: A new #-lactamase inhibitor

Markus Furegati, Sandro Nocito, Folkert Reck, Anthony Casarez, Robert Simmons, Heiner Schuetz, and Guido Koch

Org. Process Res. Dev., **Just Accepted Manuscript** • DOI: 10.1021/acs.oprd.0c00069 • Publication Date (Web): 14 May 2020

is published by the American Chemical Society. 1155 Sixteenth Street N.W., Washington, DC 20036

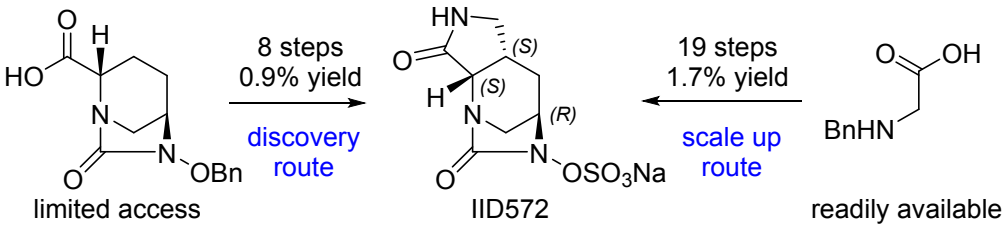
Scale up synthesis of IID572: A new β-lactamase inhibitor

Markus Furegati†, Sandro Nocito†, Folkert Reck‡, Anthony Casarez‡, Robert Simmons‡, Heiner Schuetz†, Guido Koch†*

† Synthesis and Technologies Group, Novartis Institutes for BioMedical Research, Klybeckstrasse 141, Basel 4057, Switzerland

‡ Novartis Institutes for BioMedical Research, 5300 Chiron Way, Emeryville, California 94608, United States

For Table of Contents Only:



ABSTRACT

The new potentially best-in-class β-lactamase inhibitor IID572 was discovered by a late stage functionalization approach. An alternative synthesis was developed to satisfy the short-term material need for toxicological studies in animals. The new synthetic strategy was built on two key features, an intramolecular azomethine ylide [3+2] cycloaddition that allowed the efficient formation of molecular complexity from readily available starting materials and an enzymatic resolution that resulted in high optical purity of a key intermediate.

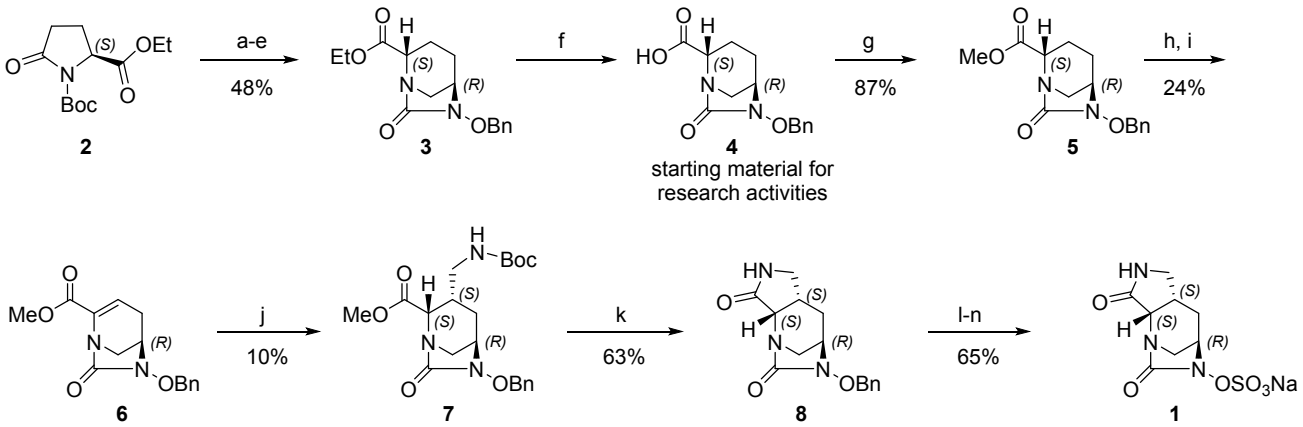
KEYWORDS

β-lactamase inhibitors, diazabicyclooctane, DBOs, scale up, enzymatic resolution, [3+2] cycloaddition

INTRODUCTION

Our research program on new β-lactamase inhibitors of the diazabicyclooctane (DBO) class revealed IID572 (**1**) to be a potent, potentially best in class, broad spectrum β-lactamase inhibitor that lacks antibacterial activity. In discovery chemistry, a late stage functionalization (LSF) approach was applied to access the unique structural framework of IID572.[1] The starting point of the research synthesis was intermediate **4** of the Avibactam synthesis (scheme 1), which can be obtained in 48% yield over 6 steps.[2, 3] The key step in our LSF route was a photochemical *Giese*-type radical addition to the *Michael* acceptor **6** that resulted in >80% yield a mixture of **7** and three other diastereoisomers (not shown).[4] After the isolation of the desired isomer **7** and deprotection with TFA the primary amine spontaneously cyclized under basic conditions to provide compound **8**.

Scheme 1. Synthesis of compound **1** by the addition of a photochemically generated radical.[a]



[a] Reagents and conditions: (a) Me_3SOI, KOtBu, THF, DMSO (b) $BnONH_2 \cdot HCl$, EtOAc (c) MsOH, EtOAc then aq. $KHCO_3$ (d) $NaBH_4$, propanoic acid, H_2SO_4, EtOAc then oxalic acid (e) triphosgene, picoline, $NaHCO_3$, DCM/H_2O (f) Novozyme 435 (yield given as 65% for the *in situ* carboxamide formation) (g) MeOH, EDC, HOBt, DIPEA, DCM (h) LDA, PhSeCl, THF (i) H_2O_2, AcOH, THF, H_2O (j) Boc-Gly-OH, $Ir[(dF(CF_3)ppy)_2(dtbbpy)]PF_6$, K_2HPO_4, 8W UVA fluorescence tube (k) TFA, then NEt_3 (l) H_2, Pd/C, MeOH (m) $SO_3 \cdot py$, Bu_4NHSO_4 (n) sodium 2-ethyl hexanoate.

This sequence, although relatively short and therefore ideal for LSF, suffers from three major drawbacks with respect to scale up. Firstly, PhSeCl, a toxic and expensive reagent was required to introduce the radical acceptor olefin and yielded **6** in only 24%. Secondly, the formation of a diastereoisomeric mixture after the radical addition resulted in formation

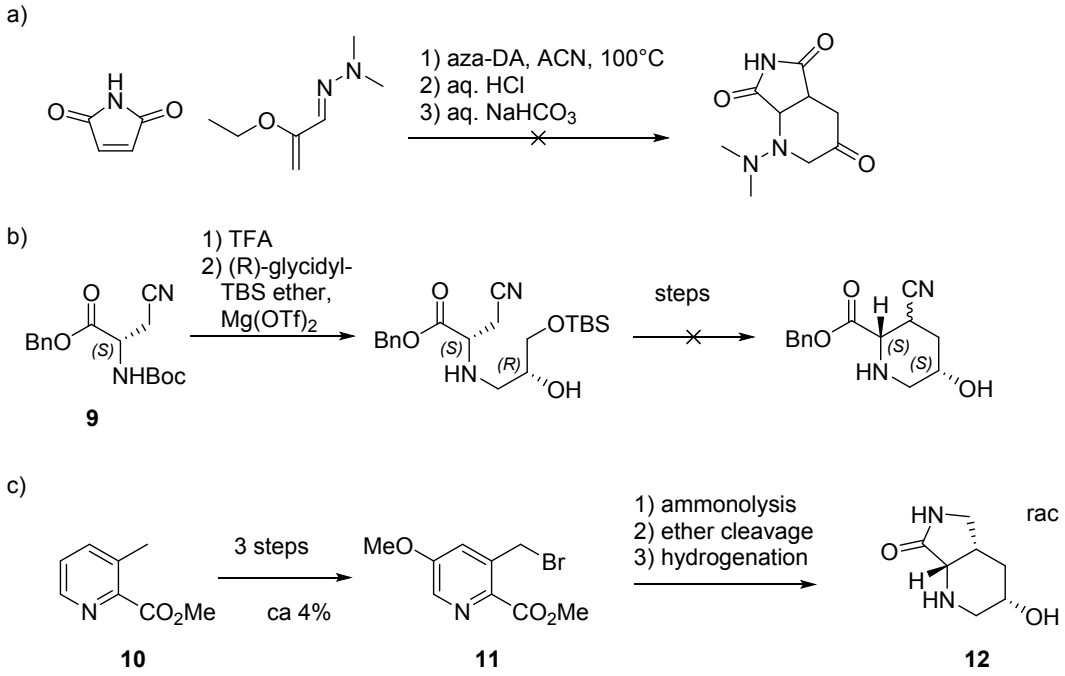
of only 10% of the desired isomer **7**. The ratio of the four isomers was not influenced by varying the reaction conditions *e.g.* solvent, catalyst or temperature. Thirdly, even if this sequence would have worked starting from the ethyl ester **3** instead of the methyl ester **5** the overall yield was calculated to be 0.48% over 12 steps. The estimated time to produce the required quantitity of the key intermediate **8** led us to investigate alternative routes with the goal to synthesize 5 g **1** as quickly as possible to enable preclinical investigation of the new development candidate.

RESULTS AND DISCUSSION

In an exploratory evaluation project we investigated a) An *Aza-Diels-Alder* route (Scheme 2a):[5] the cycloaddition, however, was a challenging key step resulting in a complex mixture of products including pyridines. b) the asparagine cyclization (Scheme 2b):[6] starting from known amino acid derived **9**.[7] The *N*-alkylation did not work well and the subsequent cyclization failed. c) Pyridine hydrogenation from known **11** (Scheme 2c):[8] Although this was a promising approach the synthesis of **11** starting from expensive methyl 3-methylpyridine-2-carboxylate (**10**) was not reproduced in a satisfactory yield; in

addition, the ammonolysis also did not work well. A similar approach was later successfully applied in an alternative Avibactam synthesis.[9] All three initial approaches were abandoned.

Scheme 2. Attempted routes

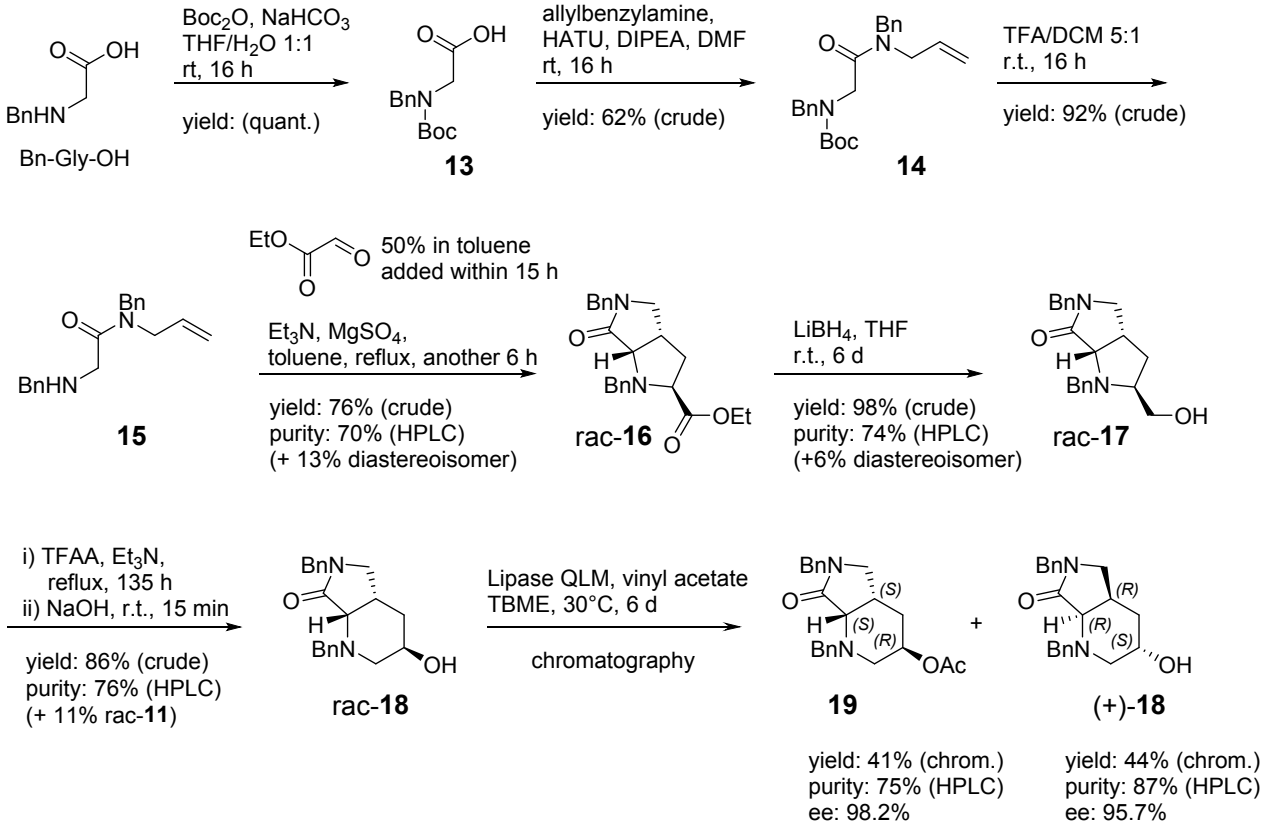


Instead, we decided to focus on a route that started from simple and easily available starting materials and allowed us to quickly build up complexity via intramolecular azomethine ylide [3+2] cycloaddition (scheme 3). The three step synthesis of **15** was described in the literature.[10] We found that Boc protection of Bn-Gly-OH[11] worked in

quantitative yield in water/THF in the presence of $NaHCO_3$. T_3P proved in our hands to be the best coupling reagent for the amide formation with commercial allylbenzylamine and deprotection with TFA gave over 3 steps an overall yield of 97% (31 g). When we outsourced this sequence the external partner delivered 1.5 kg **15** (57% yield over 3 steps using different reagents). A similar cyclization of *N*-allyl-2-(methylamino)-*N*-phenylacetamide and butyl glyoxylate was described with 30% yield.[12] We were pleased to find that slow addition of the ethyl glyoxylate significantly improved the yield of cycloaddition product rac-**16** up to 76%. The major by-product was a diastereoisomer that was difficult to remove by chromatography.[13] Reduction of the ethyl ester rac-**16** with $LiBH_4$[14]to the alcohol rac-**17** gave the starting material for the second key step, the ring expansion reaction.[15, 16] The *in situ* formed trifluoroacetate was displaced by the benzylamine in an intramolecular fashion to form an aziridinium intermediate that was re-opened at the methylene carbon (α to the ammonium) by trifluoroacetate to form the *exo* product (5-membered ring resembling the starting material) or at the higher substituted carbon to access the 6-membered ring target compound. The reaction mixture was quenched with aq. NaOH (saponification of the TFA ester) and rac-**18** was isolated

after chromatography over silica gel.[17] A long reaction time was crucial for a high yield, attempts to shorten the reaction time were not successful. Allowing the reaction to perform at higher temperature under microwave conditions resulted in higher amounts of the undesired 5-membered ring isomer and ultimately a low conversion. A single experiment applying the non-equilibrium, irreversible conditions ($MsCl/Et_3N$ followed by addition of AgOAc) gave no desired product.[15, 16] In order to avoid chiral chromatography we screened 21 lipases for stereospecific enzymatic esterification with vinyl acetate of alcohol rac-**18** and found that QLM lipase selectively provided both, the acetate and the alcohol in high enantiomeric purity (both >95% ee, E factor of 80). We completed the synthesis with one enantiomer before starting the main batch and confirmed that acetate **19** had the desired 3*R* configuration; the same as determined for the target product originally prepared by the LSF approach. While waiting for the large batch of the enzyme, we successfully reduced the amount by factor 10. The reaction proceeded smoothly and after six days the crude product was isolated and purified by the first chromatography in this sequence. Having the desired isomer at the acetate stage, an additional hydrolysis step was required to prepare key intermediate **(–)-18** (scheme 4).

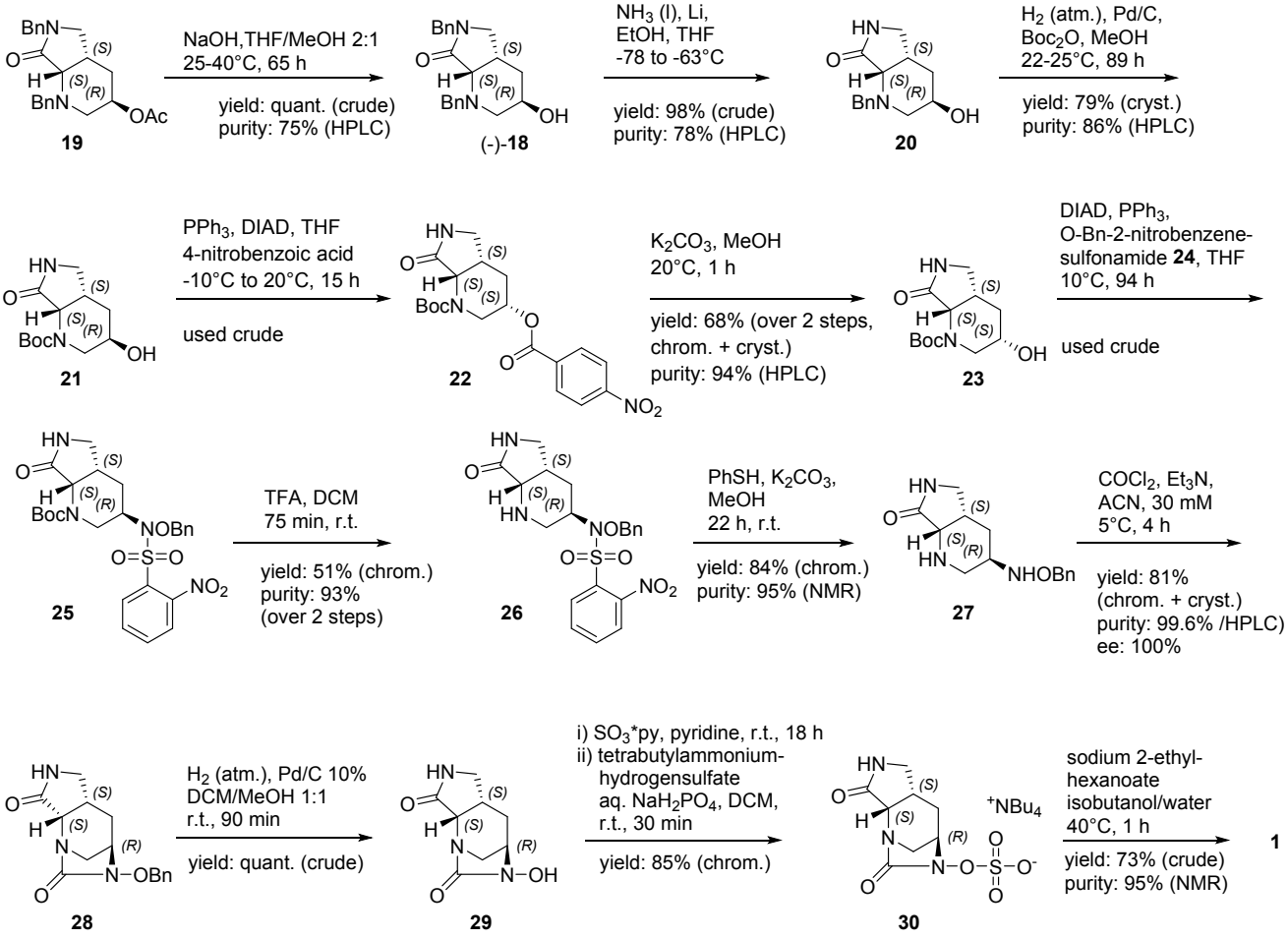
Scheme 3. Azomethine ylide route part 1: Core formation and enzymatic resolution



The end game of the synthesis dealt with protecting group manipulations, inversion of the stereo center at the 3-position, introduction of the hydroxylamine motif, urea and salt formation (scheme 4). The saponification of **19** smoothly proceeded with NaOH in THF/MeOH 2:1 in quantitative yield. The double debenzylation turned out to be more difficult than expected. After considerable optimization efforts, we chose *Birch* conditions[18] using Li and EtOH to selectively remove the benzyl group at the lactam

nitrogen first, followed by hydrogenation under standard conditions for the second benzyl cleavage. Interestingly, the two reactions also worked in the reverse order and turned out to be completely orthogonal to each other. *In situ* Boc protection during the hydrogenation step resulted in 77% yield of **21** for the sequential double debenzylation sequence. The protecting group swap from Bn to Boc was necessary for two reasons: firstly, to avoid interaction of the tertiary amine during the following *Mitsunobu* reaction and secondly, because hydrogenation was not appropriate after the introduction of the hydroxylamine motif. The inversion of the stereo center at the 3-position of **21** was accomplished by formation of the 4-nitrobenzoic ester under *Mitsunobu* conditions[19] followed by saponification with K_2CO_3 in MeOH; chromatography increased the purity to 94% by HPLC and gave **23** in 68% yield for the inversion. *Mitsunobu* condition[19] with *N*-(benzyloxy)-2-nitrobenzenesulfonamide (**24**) resulted in the formation of **25**, which after Boc deprotection and chromatography yielded **26** in 51% yield (over 2 steps).

Scheme 4. Azomethine ylide route part 2: stereo adjustments, urea and salt formation



Nosyl deprotection under standard condition[20] resulted in the formation of **27** in 84% yield. The formation of the cyclic urea was performed by slow addition of a phosgene solution (syringe pump) into a 30 mM solution of **27** and resulted in the key intermediate **28** in 81% yield after chromatography and crystallization. The chemical purity after this step was increased to 99.6% by HPLC and the ee to 100%. This material was identical with the one obtained by the original photochemical LSF approach starting from an Avibactam

precursor. In addition, an X-ray crystal structure determination of **28** confirmed its absolute stereochemistry. Debenzylation with H_2 on Pd/C gave **29** in quant. yield. Treatment with sulfurtrioxide pyridine complex followed by the addition of tetrabutylammonium hydrogensulfate in aq. sodium dihydrogenphosphate solution provided **30** in 85% yield after chromatography. Cation exchange was accomplished by treatment of **30** with sodium 2-ethylhexanoate that facilitated the undesired ion pair (e.g. the tetrabutylammonium 2-ethylhexanoate) to precipitate.[21] The target compound **1** was isolated in 73% yield and with a purity of >95% by NMR. The enantiopurity was assumed to be identical to the one measured for **28**, since we did not observe any epimerization during the transformation to **1**.

CONCLUSIONS

In order to ensure material supply for preclinical toxicological studies, we developed a *de-novo* synthesis route for the novel β-lactamase inhibitor IID572 (**1**). This was required due to a rather low yielding discovery route and unavailability of key starting materials within the necessary timelines. The herein described synthesis started from affordable,

commercially available starting materials and gave the target compound **1** between 1.7% and 2.9% overall yield in an 19-step linear sequence (total number of steps: 20, including the one step synthesis of **24**). The elegance of this synthesis is the rapid and efficient buildup of complexity from simple precursors and the application of an economic enzymatic resolution to provide the desired enantiomer in high ee. Despite some synthetic challenges with respect to functional group interconversions (scheme 4), the synthesis was successfully performed on a 8 mol scale of Bn-Gly-OH. This allowed us to deliver 10 g of **1** and significant amounts of various intermediates for further SAR optimization.

EXPERIMENTAL SECTION

General. All reagents were purchased and used as received unless otherwise noted. The larger amount of **24** was prepared according to the literature in one step.[22] Palladium on charcoal (10%) from BASF 549823. Lipase QLM (from *Alcaligenes sp*) from Meito-Sangyo Co. Ldt. Melting ranges were determined on a Leitz Biomed microscope with integrated hot plate, thermometer and an applied heat rate of 2°C/min. HPLC methods 1-3: XDB-C18 column, 4.6 mm × 50 mm, 1.8 µm, using ACN and water as eluent (both containing

0.05% TFA), column temperature of 35°C, flow rate of 1.0 mL/min, detection at 215 nm, purities were characterized with area%; method 1: gradient from 40 to 100% ACN over 6 min, 100% ACN for 1.5 min, followed by 100 to 40% ACN over 0.5 min; method 2: gradient from 5 to 100% ACN over 6 min, 100% ACN for 1.5 min, followed by 100 to 5% ACN over 0.5 min; method 3: gradient from 30 to 100% ACN over 6 min, 100% ACN for 1.5 min, followed by 100 to 30% ACN over 0.5 min. Chiral HPLC method 1: Chiralpak IC, 4.6 mm × 250 mm, 5 µm, heptane/EtOH/diethylamine 92:8:0.05, column temperature: rt, flow 1 mL/min, detection at 220 nm; method 2: Chiralpak IC, 4.6 mm × 250 mm, 5 µm, heptane/EtOH 9:1, column temperature: rt, flow 1 mL/min, detection at 210 nm; method 3: Chiralpak AD-H, 4.6 mm × 250 mm, 5 µm, heptane/EtOH 1:1, column temperature: rt, flow 1 mL/min, detection at 220 nm. LCMS methods 1 and 2: Acquity HSS T3 1.8 µm 2.1 × 50 mm column at 50°C. Eluent A: water + 0.05% formic acid + 3.75 mM ammonium acetate; eluent B: ACN + 0.04% formic acid, detection at 210-450 nm, purities were characterized with area%; method 1: gradient from 2 to 98% B in 1.4 min with a flow rate of 1.0 mL/min; method 2: gradient from 1 to 98% B (concave) in 1.4 min with a flow rate of 1.2 mL/min; method 3: Water Acquity SDS, Kinetex C18 2.6 µm 2.1 × 50 mm column

at 50°C. Eluent A: water + 0.1% TFA; eluent B: ACN + 0.1% TFA, detection at 220 nm, purities were characterized with area%; gradient from 2 to 88% B in 1.29 min and from 88% B to 95% B in 0.16 min, flow rate: 1.2 mL/min. LCMS method 4: Acquity HSS T3 1.8 µm 2.1 × 50 mm column at 60°C. Eluent A: water + 0.05% formic acid + 3.75 mM ammonium acetate; eluent B: ACN + 0.04% formic acid, detection at 210-450 nm, purities were characterized with area%; gradient from 5 to 98% B in 1.4 min with a flow rate of 1.0 mL/min. NMR spectra were recorded on a *Bruker BioSpin* machine. ^{1}H shifts were referenced to DMSO-d_6 at 2.49 ppm and $CDCl_3$ at 7.26 ppm. ^{13}C shifts were referenced to DMSO-d_6 at 39.52 ppm. LC-HRMS: The analyses was performed by using electrospray ionization in positive ion mode after separation by liquid chromatography (Vanquish, Thermo). The elemental composition was derived from the mass spectra acquired at the high resolution of about 240'000 on an Orbitrap Fusion Lumos mass spectrometer (Thermo Scientific). The high mass accuracy below <1 ppm was obtained by using an Internal Calibrant (IC). Optical rotations were measured for **18** on a Anton Paar MCP100 and for **1** on a Autopol IV automatic (Rudolph Research Analytical) polarimeter. A 30 L Buechi hastelloy reactor CR30 equipped with Huber thermostat 1015W, Flexy ALR with

automated temperature control, nitrogen inlet and dosage control. 10L, 20 L and 30 L triple jacketed AMSI Glas reactors equipped with a Huber Unistat 390W, automated temperature control, condenser and nitrogen inlet. 60 L Buechi reactor CR60 equipped with Huber thermostat 390W, Flexy ALR with automated temperature and dosage control, nitrogen inlet and condenser. For inertization, the reactors were evacuated twice to 0.3 bar and refilled with nitrogen (or argon) before adding flammable solvents.

rac-(2*S,3a*S**,6a*S**)-Ethyl 1,5-dibenzyl-6-oxooctahydropyrroloα3,4-b]pyrrole-2-carboxylate (rac-16)** A 60 L reactor was loaded with **15** (1.46 kg, 4.81 mol) and toluene (20 L). To the yellow solution magnesium sulfate (2.32 kg, 19.24 mol, 4 equiv), and triethylamine (0.87 L, 6.25 mol, 1.3 equiv) was added. The reaction mixture (pale yellow suspension) was heated to reflux (internal temperature ca 107°C) within 1 h. To the refluxing reaction mixture an ethyl glyoxylate solution 50% in toluene (1.18 kg, 5.77 mol, 1.2 equiv) was added within 15 h via a dosage pump. The reaction mixture (pale yellow suspension) was stirred for another 6 h at reflux (internal temperature ca 109°C). HPLC of the reaction mixture showed full conversion. The reaction mixture was cooled to 15°C

internal temperature, then water (20 L) was added (exotherm, the temperature raised to 32°C). The reaction mixture was stirred for 15 min, then transferred to a twin-70 L separation vessel and the phases were separated (good separation). The organic phase was washed with water (15 L) and brine (15 L). The aqueous layers were extracted twice with TBME (2 × 10 L). The second TBME phase (contained a lot of watery mud) was filtered through celite and washed with TBME. The water of the filtrate was separated and the combined organic phases concentrated in vacuo at 45°C to a volume of ca 6 L. This solution was dried over anhydrous sodium sulfate, filtered, concentrated in vacuo at 50°C and dried overnight at 50°C and 10 mbar to obtain 1.97 kg of a turbid brown oil rac-**16** (yield: 76% based on 70% HPLC-purity). The aqueous phases did not contain any product and were discarded. HPLC (method 1): t_R = 3.22 min (69.5%). LCMS (method 1): t_R = 1.21 min (64.6%, product), m/z 379.3 $[M+H]^+$; 1.16 min (12.4%, diastereoisomer), m/z 379.3 $[M+H]^+$; 1.05 min (toluene ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.43 – 7.22 (m, 16+5.6H), 4.43 (d, J = 14.9 Hz, 1H), 4.38 – 4.31 (m, 2H), 4.13 – 4.03 (m, 2+0.9H), 3.88 (d, J = 13.2 Hz, 1H), 3.74 (d, J = 8.9 Hz, 1H), 3.46 – 3.40 (m, 2H), 3.01 (ddd, J = 16.9, 8.4, 3.4 Hz, 1H), 2.96 (dd, J = 10.0, 2.8 Hz, 1H), 2.31 (s, 0.28H toluene), 2.13 (ddd, J =

13.0, 9.0, 2.4 Hz, 1H), 1.81 (dt, J = 13.1, 7.7 Hz, 1H), 1.66 (ddd, J = 12.6, 8.5, 6.7 Hz, 0.17H other diastereoisomer), 1.18 (t, J = 7.1 Hz, 4H), 1.04 (t, J = 7.1 Hz, 0.58H other diastereoisomer). LC-HRMS: calcd for $C_{23}H_{27}N_2O_3$ $[M+H]^+$ 379.20162, found 379.20169.

rac-(2*S,3a*S**,6a*S**)-1,5-Dibenzyl-2-(hydroxymethyl)hexahydropyrrolo[3,4-b]pyrrol-6(1*H*)-one (rac-17).** A 30 L hastelloy reactor was loaded with crude rac-**16** (1.97 kg, 3.64 mol, 70% purity) and THF (20 L). The brown solution was cooled to 0°C then $LiBH_4$ (238 g, 10.4 mol, 2.9 equiv) was added in portions over a period of 10 min (slightly exotherm). The reaction mixture was warmed to 23°C. After 4 d another portion of $LiBH_4$ (25 g, 1.1 mol, 0.3 equiv) was added. After another day additional $LiBH_4$ (17 g, 0.8 mol, 0.2 equiv) was added. After 6 d the conversion was complete. The reaction mixture was cooled to –10°C. 2 N hydrochloric acid (ca 8 L, 16 mol, 4.4 equiv) was added dropwise via dosage pump within two hours until pH 3 was reached (CAUTION: strong gas and foam evolution was observed!). The solution became temporarily thick and a yellow suspension was formed. After the addition the reaction mixture was stirred for 30 min at 0°C then sat. sodium bicarbonate solution (10 L) was added. The reaction mixture was transferred to

the 70 L-separation vessels and extracted with TBME (1 × 20 L and 2 × 7 L) and water (ca 8 L). The combined org. phases were washed twice with brine (2 × 10 L) and concentrated in vacuo at 45°C to a volume of ca 8 L. The residue was dried over anhydrous sodium sulfate (overnight). The suspension was filtered, the filter cake washed with TBME and the filtrate concentrated in vacuo at 50°C. The residue was dissolved in toluene (3 L), evaporated again and dried (ca 3 h) at 50°C and 10 mbar to obtain 1630 g of a turbid yellow-brown oil rac-**17** (yield: 98% based on 74% HPLC-purity). HPLC (method 2): t_R = 3.64 min (74.1%). LCMS (method 1): t_R = 0.77 min (73.9%, product), m/z 337.3 $[M+H]^+$; 0.80 min (6.7%, diastereoisomer), m/z 337.3 $[M+H]^+$; 1.05 min (toluene). The crude material contained about 7% of the diastereoisomer and 3% toluene and was used without further purification. A small sample was purified for NMR: ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.44 – 7.19 (m, 10H), 4.52 – 4.45 (m, 2H), 4.29 (dd, J = 36.5, 14.3 Hz, 2H), 4.08 (d, J = 13.7 Hz, 1H), 3.58 (d, J = 8.1 Hz, 1H), 3.45 (ddd, J = 10.9, 5.5, 4.0 Hz, 1H), 3.39 – 3.32 (m, 2H?+H_2O signal), 2.92 (dd, J = 9.9, 2.3 Hz, 1H), 2.87 (qd, J = 6.3, 3.9 Hz, 1H), 2.76 (dtdd, J = 10.0, 7.6, 5.1, 2.2 Hz, 1H), 2.00 – 1.94 (m, 1H), 1.60 (ddd, J = 12.6, 7.2, 5.1 Hz, 1H), 1.18 (t, J = 7.1 Hz, 5% EtOAc). LC-HRMS: calcd for $C_{21}H_{25}N_2O_2$

$[M+H]^+$ 337.19105, found 337.19104.

rac-(3*R,4a*S**,7a*S**)-1,6-Dibenzyl-3-hydroxyoctahydro-7*H*-pyrrolo[3,4-b]pyridin-7-one (rac-18)**. A 30 L hastelloy reactor was loaded with crude rac-**17** (1.63 kg, 3.59 mol, 74% purity), molecular sieves 4 Å (2.5 kg) and THF (23 L). The resulting brown solution was cooled to –5°C, then trifluoroacetic anhydride (0.820 L, 5.81 mol, 1.6 equiv) was added dropwise within 35 min. The reaction mixture was stirred for 1.5 h while warming to rt. To the reaction mixture was added triethylamine (3.37 L, 24.2 mol, 6.7 equiv) within 10 min, then heated at reflux (jacket temperature 75°C) for 135 h. During this time the ratio of the product (after hydrolysis) and its 5-membered ring isomer changed from 55:45 (after 15 h) to 90:10 (equilibrium). For the IPCs a samples of the reaction mixture was quenched with 0.5 N NaOH and checked with LCMS. The reaction mixture was cooled to rt and poured into a 70 L vessel containing 1 N aq. NaOH solution (24 L, 24 mol, 6.7 equiv) and stirred for 15 min. To the resulting brown suspension was added celite (3 kg) and the suspension stirred for 15 min after which it was filtered over a pad of celite to remove of the molecular sieves and washed with TBME (filtration took ca 5 h). To the filtrate was

added TBME (20 L) and after extraction and phase separation the aq. phase was extracted with TBME (2 × 7 L). The combined org. phases were washed with sat. sodium bicarbonate solution (10 L) and brine (15 L) and concentrated in vacuo at 45°C to a volume of ca 8 L. After drying with anhydrous sodium sulfate (2 kg) the suspension was filtered through silica gel (40-63 µm, 1 kg) to remove baseline impurities. The silica gel bed was washed with EtOAc (4 × 2 L) until no product eluted. The filtrate was concentrated in vacuo at 45°C and dried 3 h at 50°C and 15 mbar to obtain 1366 g rac-**18** as a dark brown oil, which was used crude for the next step (yield: 86% based on 76% HPLC-purity (the rest was mostly sm)). HPLC (method 2): t_R = 3.70 min (75.8%). LCMS (method 1): t_R = 0.84 min (65.9%, product), m/z 337.3 $[M+H]^+$; 0.77 min (9.9%, starting material) ES+ m/z 337.3 $[M+H]^+$; 0.86 min (7.7%, diastereoisomer), m/z 337.3 $[M+H]^+$. A small sample was purified for NMR: ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.39 – 7.21 (m, 10H), 4.67 – 4.59 (m, 2H), 4.43 (d, J = 13.9 Hz, 1H), 4.25 (d, J = 15.1 Hz, 1H), 3.54 (tq, J = 9.1, 4.3 Hz, 1H), 3.41 (d, J = 13.9 Hz, 1H), 3.18 (d, J = 8.2 Hz, 2H), 2.85 (d, J = 5.7 Hz, 1H), 2.70 (ddd, J = 10.9, 3.9, 1.4 Hz, 1H), 2.61 (ddt, J = 8.5, 5.8, 2.8 Hz, 1H), 1.78 – 1.68 (m, 2H), 1.34 (ddd, J = 13.4, 10.0, 6.1 Hz, 1H). LC-HRMS: calcd for $C_{21}H_{25}N_2O_2$ $[M+H]^+$

337.19105, found 337.19113.

(3*R*,4a*S*,7a*S*)-1,6-Dibenzyl-7-oxooctahydro-1*H*-pyrrolo[3,4-b]pyridin-3-yl acetate (19). A 30 L glass reactor was loaded with crude rac-**18** (1.36 kg, 3.05 mol, 76% purity), TBME (21 L), vinyl acetate (4.20 L, 45.6 mol, 15 equiv) and lipase QLM (25 g, activity: 101400 U/g). The suspension was stirred (200 rpm) at 30°C internal temperature. After 143 h HPLC the reaction mixture showed a 50:50 mixture of alcohol and acetate. The reaction mixture was cooled to 20°C and filtered over celite (0.5 kg). The filtrate was concentrated in vacuo at 35°C to a volume of 3 L then toluene (1 L) was added and concentrated in vacuo first at 35°C then at 50°C to dryness. 1689 g of a dark brown honey was obtained. This crude product was dissolved in TBME / heptane 2:1 (4.4 L) and purified in 14 portions (12 + 2 mixed fractions) by column chromatography. The same batch of silica gel (7 kg, 25-40 µm, 60Å, Macherey-Nagel) in an axially compressed chromatography column was used for the entire purification; a ternary gradient (heptane/EtOAc/MeOH) was applied, followed by a wash program (1/2 column volume MeOH) and re-conditioning, flow rate 1 L/min. The purification resulted in 626 g brown honey **19** (yield: 41% based on 75%

HPLC-purity). HPLC (method 3): t_R = 2.47 min (75.0%). Chiral HPLC (method 1): 98.2%ee, t_R (3*S*,4a*R*,7a*R*-enantiomer) = 23.95 min, t_R (3*R*,4a*S*,7a*S*-enantiomer 2) = 33.75 min. LCMS (method 1): t_R = 1.16 min (75.5%, product), m/z 379.3 $[M+H]^+$; 1.15 min (15.6%, 5-membered ring isomer (signal at 3.62 ppm)), m/z 379.3 $[M+H]^+$. ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.47 – 7.15 (m, 10+4H), 4.72 (qd, J = 6.0, 3.1 Hz, 1H), 4.55 (d, J = 14.9 Hz, 1H), 4.32 (dd, J = 38.6, 14.5 Hz, 2H), 3.75 (d, J = 14.3 Hz, 1H), 3.29 – 3.21 (m, 2H), 3.00 (dd, J = 9.6, 5.8 Hz, 1H), 2.71 (dd, J = 11.6, 3.2 Hz, 1H), 2.62 (h, J = 6.2 Hz, 1H), 2.23 (dd, J = 11.6, 7.1 Hz, 1H), 1.97 (s, 3H), 1.66 (t, J = 6.1 Hz, 2H). LC-HRMS: calcd for $C_{23}H_{27}N_2O_3$ $[M+H]^+$ 379.20162, found 379.20172.

(3*S*,4a*R*,7a*R*)-1,6-Dibenzyl-3-hydroxyoctahydro-7*H*-pyrrolo[3,4-b]pyridin-7-one ((+)-18)

From the previously described chromatography 510 g of the undesired enantiomer (alcohol form) as a dark brown honey was collected from chromatography runs 1-12 **(+)-18** (yield: 44% based on 87% HPLC-purity). $[\alpha]_D^{25}$ +59.6 (c=1.0 w/v%, $CHCl_3$). HPLC (method 3): t_R = 1.58 min (86.7%). LCMS (method 1): t_R =0.85 min (85.6%, product), m/z 337.3 $[M+H]^+$; 0.87 min (9.7%, 5-membered ring isomer), m/z 337.3 $[M+H]^+$. Chiral HPLC

(method 2): 95.7%ee, t_R (3R,4aS,7aS) = 15.98 min, t_R (3S,4aR,7aR) = 16.94 min. ^{1}H NMR (600 MHz, DMSO-d6) δ ppm 1.34 (ddd, J=13.43, 9.95, 6.14 Hz, 1 H) 1.69 - 1.79 (m, 2 H) 2.56 - 2.65 (m, 1 H) 2.70 (dd, J=10.36, 3.03 Hz, 1 H) 2.86 (d, J=5.87 Hz, 1 H) 3.18 (d, J=8.25 Hz, 2 H) 3.42 (d, J=13.75 Hz, 1 H) 3.50 - 3.60 (m, 1 H) 4.21 - 4.30 (m, 1 H) 4.43 (d, J=13.94 Hz, 1 H) 4.57 - 4.65 (m, 2 H) 7.16 - 7.44 (m, 12 H) contained 2% residual solvent. LC-HRMS: calcd for $C_{21}H_{25}N_2O_2$ $[M+H]^+$ 337.19105, found 337.19107.

(3*R*,4a*S*,7a*S*)-1,6-Dibenzyl-3-hydroxyoctahydro-7*H*-pyrrolo[3,4-b]pyridin-7-one ((–)-18). A 20 L flask of a rotavapor was evacuated twice to 0.3 bar and re-filled with nitrogen and loaded with **19** (616 g, 1.22 mol, 75% purity), THF (4 L) and aq. 2 N NaOH (3.97 L, 7.94 mol, 6.5 equiv). The biphasic mixture was vigorously stirred for 18 h at 25°C and for another 7 h at 40°C in the rotavapor water bath. IPC (HPLC) showed 19% sm. MeOH (2 L) was added and the reaction mixture was stirred for 18 h at 25°C, 7 h at 40°C and another 15 h at rt, then HPLC indicated full conversion. The reaction mixture was extracted with TBME (1 × 6 L and 3 × 3 L). The combined organic phases were washed with brine (4 L) and concentrated in vacuo at 45°C to a volume of 5 L. Anhydrous sodium

sulfate (1 kg) was added, the suspension filtered and the filtrate concentrated in vacuo at 45°C. The residue was re-dissolved in toluene (3 L) and concentrated again and dried for 2 h at 60°C and 20 mbar to obtain 555 g of a brown honey **(–)-18** (yield: quant. based on 76% HPLC-purity). $[\alpha]_D^{25}$ –56.1 (c=1.0 w/v%, $CHCl_3$). HPLC (method 3): t_R = 1.58 min (76.4%). LCMS (method 1): t_R = 0.78 min (14.0%, 5-membered ring isomer), ES+ m/z 337.3 $[M+H]^+$; 0.84 min (74.3%, product), ES+ m/z 337.3 $[M+H]^+$. ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.39 – 7.19 (m, 13H), 4.65 – 4.59 (m, 2H), 4.42 (d, J = 13.9 Hz, 1H), 4.32 (d, J = 13.7 Hz, 0.28H (main impurity)), 4.25 (dd, J = 15.0, 3.0 Hz, 1H), 3.54 (tq, J = 9.1, 4.4 Hz, 1H), 3.42 (d, J = 13.9 Hz, 1H), 3.18 (d, J = 8.2 Hz, 2H), 2.85 (d, J = 5.8 Hz, 1H), 2.70 (ddd, J = 10.8, 3.8, 1.4 Hz, 1H), 2.65 – 2.56 (m, 1H), 1.78 – 1.70 (m, 2H), 1.34 (ddd, J = 13.4, 9.9, 6.1 Hz, 1H). LC-HRMS: calcd for $C_{21}H_{25}N_2O_2$ $[M+H]^+$ 337.19105, found 337.19104.

(3*R*,4a*S*,7a*S*)-1-Benzyl-3-hydroxyoctahydro-7*H*-pyrrolo[3,4-b]pyridin-7-one (20)

Equipment: 30 L hastelloy reactor with an ammonia inlet and an argon inlet attached to a gas scrubber filled with 100 L aq. 30% sulfuric acid solution. The reactor was cooled to –

80°C and loaded with liquid ammonia (12.7 L, 587 mol) (note: the gas cylinders were pre-cooled in dry ice for about 10 min), a solution of **(–)-18** (543 g, 1.21 mol, purity 76%) in THF (1.5 L) and anhydrous EtOH (236 mL, 4.04 mol, 3.3 equiv). Finally, granular lithium (99% trace metals basis, 44.8 g, 6.46 mol, 5.3 equiv) was added portion wise within 15 min (temperature raised from –72°C to –63°C). After 5 min the color changed from yellow to gray to dark blue. The reaction mixture (suspension) was stirred at –65°C internal temperature. After 10 min, the color changed back to gray. 1st IPC (HPLC) indicated 45% remaining starting material. To the reaction mixture lithium (22.4 g, 3.23 mol, 2.7 equiv) were added at –63°C (temperature raised to –56°C, the color changed immediately to dark blue). To the reaction mixture anhydrous EtOH (94 mL, 1.62 mol, 1.3 equiv) was added and stirred at –60°C. The color changed to gray after 30 min. 2nd IPC (HPLC) indicated 20% remaining starting material. Lithium (11.2 g, 1.62 mol, 1.3 equiv) and anhydrous EtOH (47 mL, 0.81 mol, 0.7 equiv) was added at –60°C (blue color). After 45 min anhydrous EtOH (47 mL, 0.81 mol, 0.7 equiv) was added at –60°C and after further 10 min the color turned gray. 3rd IPC (HPLC) indicated 10% remaining starting material. Lithium (11.2 g, 1.62 mol, 1.3 equiv) was added, the color remained blue after stirring

overnight at –60°C. Anhydrous EtOH (94 mL, 1.62 mol, 1.3 equiv) was added, the color turned gray after 15 min. 4th IPC (HPLC) indicated 3-5% remaining starting material. The reaction mixture was quenched by the addition of solid ammonium chloride (2003 g, 37.4 mol, 31 equiv) portion wise within 10 min (temperature raised to –42°C). The reaction mixture was warmed (–28°C jacket temperature) and stirred overnight, then the jacked temperature was raised to –2°C to allow the complete evaporation of ammonia that was all neutralized in the gas scrubber. To the residue water (15 L) and TBME (8 L) was added (pH 12-13), followed by aq HCl 32% until pH 9-10 was obtained (product is soluble in water at pH 12-13) and extracted. The aqueous phase was washed with DCM (3 × 2 L). The combined organic phases were washed with brine (5 L) and concentrated in vacuo at 45°C to a volume of ca 3 L. The residue was dried over anhydrous sodium sulfate (1 kg). The suspension was filtered and the filtrate was concentrated in vacuo at 45°C. Then dried for 2 h at 65°C and 20 mbar to deliver 373 g of a brown resin **20** (yield: 98% based on 78% HPLC-purity). HPLC (method 2): t_R = 2.17 min (78.5%). LCMS (method 2): t_R = 0.75 min (ca 87%, product), ES+ m/z 247 $[M+H]^+$. A small sample was purified for NMR: ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.79 (s, 1H), 7.40 – 7.16 (m, 5H), 4.62 (d, J = 4.9 Hz,

1H), 4.32 (d, J = 13.9 Hz, 1H), 3.62 (tq, J = 9.2, 4.4 Hz, 1H), 3.43 (d, J = 13.9 Hz, 1H), 3.13 (d, J = 8.1 Hz, 2H), 2.69 (ddd, J = 10.7, 3.9, 1.4 Hz, 1H), 2.64 (d, J = 6.0 Hz, 1H), 2.57 (ttd, J = 8.5, 5.9, 3.2 Hz, 1H), 1.73 (ddd, J = 19.8, 9.7, 6.5 Hz, 2H), 1.34 (ddd, J = 13.3, 9.9, 6.1 Hz, 1H). LC-HRMS: calcd for $C_{14}H_{19}N_2O_2$ $[M+H]^+$ 247.14410, found 247.14412.

tert-Butyl (3*R*,4a*S*,7a*S*)-3-hydroxy-7-oxooctahydro-1*H*-pyrrolo[3,4-b]pyridine-1-carboxylate (21) To a solution of **20** (372 g, 1.18 mol, purity 78%) and Boc_2O (270 g, 1.24 mol, 1.05 equiv) in THF (4 L) was added palladium on charcoal 10% (15 g, 0.014 mol, 0.01 equiv) and the reaction mixture hydrogenated at 0.1 bar and 22-25°C in a shaking duck reactor. After 18 h (57% hydrogen consumption) another portion of palladium on charcoal 10% (15 g, 0.014 mol, 0.01 equiv) was added. After total 89 h complete conversion was observed by HPLC. The reaction mixture was filtered through celite, washed with THF and concentrated in vacuo to obtain 545 g (wet) of the crude product as a pale brown solid. The crude product was suspended in EtOAc (1 L) and stirred well for 1 h at 75°C bath temperature. To the pale brown suspension heptane (1.5 L) was

slowly added at 75°C and the suspension stirred for 2 h at rt. The product was filtered off and the solid was washed with heptane in several portions. The solid was dried in vacuo at 45°C until constant weight was obtained: 278.5 g **21** as white crystals (yield: 79% based on 86% HPLC-purity). Melting range: 172-174°C. HPLC (method 2): t_R = 2.55 min (86.1%). LCMS (method 2): t_R = 0.86 min (ca 70%, product), ES+ m/z 257.3 $[M+H]^+$. ^1H NMR (600 MHz, DMSO-d_6) δ 7.72 (d, J = 21.2 Hz, 1H), 4.80 – 4.49 (m, 2H), 3.76 (d, J = 45.5 Hz, 2H), 3.37 (s, 1H), 2.72 (d, J = 9.8 Hz, 1H), 2.60 – 2.44 (m, 2H), 1.82 – 1.71 (m, 1H), 1.47 – 1.36 (m, 9H), 1.31 (q, J = 12.9, 11.1 Hz, 1H). LC-HRMS: calcd for $C_{12}H_{21}N_2O_4$ $[M+H]^+$ 257.14958, found 257.14960.The mother liquor was concentrated in vacuo at 45°C to obtain 162 g as a brown oil that contained <5% product according to HPLC and was discarded.

tert-Butyl (3*S*,4a*S*,7a*S*)-3-((4-nitrobenzoyl)oxy)-7-oxooctahydro-1*H*-pyrrolo[3,4-b]pyridine-1-carboxylate (22) Equipment: 20 L glass reactor. A pale brown suspension of **21** (270 g, 0.91 mol, purity 86%), in THF (13 L) was cooled to –10°C. 4-nitrobenzoic acid (323 g, 1.90 mol, 2.1 equiv) and triphenylphosphine (524 g, 1.90 mol, 2.1 equiv) were

added at –5°C internal temperature. Then a solution of diisopropyl azodicarboxylate (359 ml, 1.90 mol, 2.1 equiv) in THF (1.3 L) was added dropwise within 30 min at –4 to –10°C (set outside temperature to –10°C). After the addition the reaction mixture was allowed to warm to room temperature (set outside temperature to 20°C) and stirred overnight. The reaction mixture turned into a pale yellow solution. IPC after 15 h (HPLC) showed full conversion. The reaction mixture was concentrated in vacuo at 45°C to give 1643 g (wet) crude product as a brown oily resin that was immediately used for the next step. HPLC (method 2): t_R = 3.56 min (6.7%, 4-nitrobenzoic acid); 4.57 min (75.8%, P(O)Ph3); 4.77 min (8.6%, 4-nitrobenzoate target intermediate).

tert-Butyl (3*S*,4a*S*,7a*S*)-3-hydroxy-7-oxooctahydro-1*H*-pyrrolo[3,4-b]pyridine-1-carboxylate (23) The crude 4-nitrobenzoate ester (**22**) from the previous step was suspended in MeOH (15 L) then potassium carbonate (393 g, 2.84 mol, 3.1 equiv) was added. The resulting yellow suspension was stirred at room temperature for 1 h. IPC (HPLC) showed full conversion after 45 min. The reaction mixture was concentrated in vacuo at 40°C to give an orange solid residue. DCM (6 L) was added and the orange

suspension stirred for 30 min at rt. The suspension was filtered (HPLC of the filter cake showed only 4-nitrobenzoic acid as the potassium salt), washed with DCM and the filtrate was concentrated in vacuo at 45°C to give 1435 g of crude product as an orange brown oily solid. This crude product was suspended in DCM/MeOH 97:3 (4 L), stirred for 30 min at rt, filtered and washed with DCM (HPLC of the filter cake revealed again no product). The filtrate was concentrated in vacuo at 45°C to a volume of 3 L and purified in two portions by column chromatography on silica gel (7 kg, 25-40 µm, 60Å, Macherey-Nagel). The same batch of silica gel in an axially compressed chromatography column was used for the entire purification; a binary gradient (DCM/MeOH) was applied, followed by a wash program (1/2 column volume MeOH) and re-conditioning, flow rate 1L/min. The purification resulted in 189 g product as a pale brown solid. According to ^{1}H-NMR this product still contained 1% of 4-nitrobenzoic acid and 4-5% of triphenylphosphine oxide. Crystallization: This product was dissolved in DCM/MeOH 95:5 (5 L) at 45°C bath temperature. To the clear brown solution heptane (5 L) was added slowly, no crystallization. DCM was distilled off in vacuo at 45°C bath temperature (solvent exchange) the product started to crystallize after ca 15 min. The beige suspension was

stirred for about 1 h at rt, then filtered off and the solid washed with heptane. The solid was dried in vacuo at 45°C until constant weight was obtained. 167.7 g of product as pale brown crystals was obtained **23** (yield: 68% (over 2 steps) based on 94% HPLC-purity). Melting range: 223-224°C. HPLC (method 2): t_R = 2.90 min (94.2%). LCMS (method 2): t_R = 0.95 min (main signal, product) ES+ m/z 257.3 $[M+H]^+$. ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.80 (d, J = 20.0 Hz, 1H), 4.97 (t, J = 4.6 Hz, 1H), 4.66 (d, J = 7.0 Hz, 0.5H (rotamers)), 4.51 (d, J = 7.1 Hz, 0.5H (rotamers)), 3.91 (ddd, J = 22.3, 13.1, 4.3 Hz, 1H), 3.37 (ddd, J = 9.9, 5.2, 2.2 Hz, 1H), 3.33 – 3.27 (m, 1H), 2.77 (ddd, J = 9.8, 4.6, 2.0 Hz, 1H), 2.45 (ddq, J = 17.9, 12.1, 5.9 Hz, 1H), 2.17 (dd, J = 12.5, 10.6 Hz, 0.5H (rotamers)), 2.02 (dd, J = 12.4, 10.6 Hz, 0.5H (rotamers)), 1.99 – 1.91 (m, 1H), 1.43 (s, 4.5H (rotamers)), 1.39 (s, 4.5H (rotamers)), 1.06 (dq, J = 15.2, 12.1 Hz, 1H). LC-HRMS: calcd for $C_{12}H_{21}N_2O_4$ $[M+H]^+$ 257.14958, found 257.14957. The mother liquor was concentrated in vacuo at 45°C to yield 18 g of a brown resin that was discarded (HPLC showed mainly 4-nitrobenzoic acid and triphenylphosphine oxide).

tert-Butyl (3*R*,4a*S*,7a*S*)-3-((*N*-(benzyloxy)-2-nitrophenyl)sulfonamido)-7-oxooctahydro-

1*H*-pyrrolo[3,4-b]pyridine-1-carboxylate (25) Equipment: 10 L glass reactor with an additional 1 L dropping funnel. A pale brown suspension of **23** (100 g, 367 mmol, purity 94%) in THF (6 L) was cooled to –14°C internal temperature. Triphenylphospine (124 g, 473 mmol, 1.3 equiv) and N-(benzyloxy)-2-nitrobenzenesulfonamide **(24)** [22] (140 g, 454 mmol, 1.2 equiv) were added. Then a solution of diisopropyl azodicarboxylate (88 ml, 454 mmol, 1.2 equiv) in THF (0.6 L) was added dropwise within 50 min. After the addition the reaction mixture was warmed to 10°C internal temperature within 2.5 h. The reaction mixture went into solution after about 1 h and then turned again to a suspension after 2 h at 10°C and was further stirred for 91 h when IPC (HPLC) showed full conversion. The reaction mixture was concentrated in vacuo at 45°C to give 495 g of product as a brown oily resin. HPLC (method 2): t_R = 5.31 min (16.0%). LCMS (method 1): t_R = 0.96 min (39.1%, $P(O)Ph_3$), m/z 279 $[M+H]^+$; 1.05 min (27.5%, sulfonamide sm), m/z 307 $[M+H]^-$; 1.13 min (21.6%, product), m/z 547 $[M+H]^+$; 1.30 min (2.8%, isopropylcarbamate of product), m/z 633 $[M+H]^+$; 1.44 min (4.8%, PPh_3), m/z 263 $[M+H]^+$. LC-HRMS: calcd for $C_{25}H_{31}N_4O_8S$ $[M+H]^+$ 547.18571, found 547.18585. The crude product **25** was used crude in the next step.

_N_-(Benzyloxy)-2-nitro-_N_-((3_R_,4a_S_,7a_S_)-7-oxooctahydro-1_H_-pyrrolo[3,4-b]pyridin-3-yl)benzenesulfonamide (26) Equipment: 10 L glass reactor. To a pale brown solution of **25** (495 g, 367 mmol, crude from last step) in DCM (5 L) TFA (990 ml, 12.85 mol, 35 equiv) was added. The reaction mixture was stirred for 75 min at rt when IPC (HPLC) showed full conversion. The reaction mixture was concentrated in vacuo at 40°C. To the residue was added DCM (3 × 1 L) and the solvent was removed in vacuo. This resulted in the formation of 865 g crude product as the TFA salt in form of a brown oily solid, which was dissolved in MeOH (8 L) and cooled to 10°C. To this brown solution was added potassium carbonate (784 g, 5.67 mol, 15.4 equiv) in several portions (formation of gas was observed). The brown suspension was stirred for 60 min at 20°C internal temperature. IPC (HPLC) showed no isopropylcarbamate by-product. To the reaction mixture was added celite (0.5 kg) and the suspension stirred for 5 min, filtered over celite and the filter cake washed with DCM (5 L). The yellow filtrate (only MeOH part) was acidified with aq. HCl 32% (ca 400 mL) until pH 4-5 was reached. The pale brown suspension was concentrated in vacuo at 40°C. The residue was combined with the DCM filtrate (5 L) and extracted with sat. sodium bicarbonate solution (2 × 3 L). The aqueous

layer was washed with DCM (3 × 2 L). The combined organic phases were dried over anhydrous sodium sulfate, filtered and concentrated in vacuo at 45°C to give 444 g of crude product as a pale brown oily resin, which was dissolved in DCM (1 L) and purified in one portion by column chromatography on silica gel (7 kg) using a ternary gradient (DCM/EtOAc/MeOH), followed by a wash program (1/2 column volume MeOH) and re-conditioning, flow rate 800 mL/min. The purification resulted in 83.8 g product as a pale brown solid **26**. Melting range: 164-168°C. HPLC (method 2): t_R = 3.89 min (92.6%). LCMS (method 1): t_R = 0.82 min (89.7%, product), m/z 447.3 $[M+H]^+$. ^{1}H NMR (600 MHz, DMSO-d_6) δ 8.09 (dd, J = 8.0, 1.3 Hz, 1H), 8.06 (dd, J = 8.0, 1.3 Hz, 1H), 8.01 (td, J = 7.7, 1.4 Hz, 1H), 7.89 (td, J = 7.7, 1.3 Hz, 1H), 7.68 – 7.52 (m, 1H), 7.42 (d, J = 2.1 Hz, 5H), 5.00 (s, 2H), 3.78 (s, 1H), 3.16 – 2.97 (m, 3H), 2.82 (d, J = 95.4 Hz, 1H), 2.58 (dd, J = 11.5, 8.4 Hz, 2H), 2.40 (s, 1H), 1.67 (d, J = 62.5 Hz, 1H), 1 proton missing, may be under 7.6 ppm. LC-HRMS: calcd for $C_{20}H_{23}N_4O_6S$ $[M+H]^+$ 447.13328, found 447.13333. Mixed fractions were purified via chromatography on a 120 g column eluting with EtOAc 100% then with DCM/MeOH 98:2 to 85:15. Another 5.21 g **25** were isolated as a pale brown solid. HPLC (method 2): t_R = 3.89 min (94.8%). LCMS (method 1): t_R = 0.82 min

(89.7%, product), m/z 447.3 $[M+H]^+$. HPLC (method 2): t_R = 2.90 min (94.2%). Total 89.0 g of product was obtained (yield over 2 steps: 51% based on 93% calculated HPLC-purity).

(3*R*,4a*S*,7a*S*)-3-((Benzyloxy)amino)octahydro-7*H*-pyrrolo[3,4-b]pyridin-7-one (27). Equipment: 4.5 L 5-neck reaction flask with mechanical stirrer, internal thermometer, 250 mL dropping funnel, condenser, nitrogen inlet and gas scrubber filled with bleach solution (10%). A pale brown suspension of **26** (87.0 g, 181 mmol, purity 93%) in ACN (2 L) was cooled in an ice bath to 5°C. Potassium carbonate (128 g, 926 mmol, 5.1 equiv) and thiophenol (151 ml, 1.48 mol, 8.2 equiv) were added. The cooling bath was removed and the reaction mixture stirred for 22 h at rt. The color changed from pale brown to yellow and overnight an orange brown suspension was obtained. IPC (HPLC) showed full conversion. The reaction mixture was filtered through celite, washed with EtOAc, the filtrate concentrated in vacuo at 40°C and suspended in EtOAc (1 L). The fine suspension was stirred for 15 min at rt, filtered again through celite, washed with EtOAc and the filtrate was concentrated again in vacuo at 40°C to give 220 g of crude product as a brown oil.

The crude product was dissolved in DCM (150 mL) and purified by chromatography on a 340 g silica gel column eluting with DCM/MeOH 0 to 20%. The product fractions were combined and concentrated in vacuo to result in 46.8 g of product as a brown resin (purity by HPLC: ca 79%). A second chromatography on 1.2 kg spherical silica gel (30 µm, 70 Å, Morvay) eluting with DCM/MeOH 2 to 20% resulted in 41.8 g **27** as a brown resin after being dried in vacuo at 45°C that crystallized after a few minutes to a brown hard solid (yield: 84% based on 95% NMR-purity). Melting range: 124-126°C. HPLC (method 2): t_R = 2.76 min (86.1%). LCMS (method 1): t_R = 0.46 min (66.7%, product), m/z 262.3 $[M+H]^+$. ^{1}H NMR (600 MHz, DMSO-d_6) δ 7.59 (s, 1H), 7.38 – 7.31 (m, 4H), 7.31 – 7.27 (m, 1H), 6.55 (d, J = 7.5 Hz, 1H), 4.62 (s, 2H), 3.19 – 3.12 (m, 2H), 2.97 (s, 1H), 2.88 (ddd, J = 9.4, 5.0, 1.2 Hz, 1H), 2.72 (dd, J = 11.9, 3.0 Hz, 1H), 2.45 (dd, J = 11.8, 6.1 Hz, 1H), 2.36 (h, J = 6.4 Hz, 1H), 1.59 (dt, J = 13.3, 6.5 Hz, 1H), 1.50 (ddd, J = 13.7, 7.4, 4.2 Hz, 1H), 1 proton not visible. LC-HRMS: calcd for $C_{14}H_{20}N_3O_2$ $[M+H]^+$ 262.1550, found 262.15506. Notes: The second column chromatography did not significantly improve the purity. The purity is higher in ^{1}H-NMR (ca 95%) compared to HPLC and LCMS.

(4*R*,5a*S*,8a*S*)-3-(Benzyloxy)hexahydro-2*H*-1,4-methanopyrrolo[3,4-d][1,3]diazepine-2,8(3H)-dione (28). Equipment: 6 L 5-neck reaction flask with mechanical stirrer, internal thermometer, argon inlet and ice bath. The yellow solution of **27** (31 g, 113 mmol, 95% purity) and *N*,*N*-diisopropylethylamine (59.0 ml, 338 mmol, 3 equiv) in dry ACN (3.47 L) was cooled in an ice bath to 5°C internal temperature. A solution of phosgene (20% in toluene, 62.3 ml, 118 mmol, 2.9 equiv) in ACN (495 mL) was added via syringe pump within 3 h (flow rate: 3 mL/min) (**CAUTION**: phosgene is highly toxic. The use of a gas sensor as part of the PPE is recommended) The reaction mixture was stirred for one additional hour at 5°C. IPC (HPLC) showed full conversion. The reaction mixture was concentrated in vacuo at 40°C to a volume of 400 mL. The residue was diluted with DCM (800 mL) and poured into ice-cold 0.1 N aq. HCl, saturated with NaCl (1 L, 0.1 mol HCl, 0.9 equiv). After extraction the phases were separated and the aqueous layer washed with DCM (3 × 300 mL). The combined organic phases were washed with brine (1 L) and dried over anhydrous sodium sulfate, filtered and concentrated in vacuo at 45°C to yield 39.6 g of a yellow brown solid that was dissolved in DCM/MeOH 97:3 (300 mL) and purified in two portions by column chromatography on 1.2 kg spherical silica gel (30 µm, 70 Å, Morvay)

eluting with DCM/MeOH 2 to 8%. The combined product fractions (concentrated to a volume of 1 L) contained 3-4% of an isomer. To this crude product solution was added product from a previous experiment (1.96 g). To the clear pale brown solution was slowly added EtOAc (800 ml) at 40°C. The product started to crystallize after 1 min. DCM and part of the EtOAc were distilled off in vacuo at 40°C bath temperature until a volume of 0.3 L. The pale brown suspension was stirred for 1 h at rt. The product was filtered off and the solid washed with ice cold EtOAc in three portions (3 × 80 mL). The solid was dried in vacuo at 45°C (overnight) until constant weight was obtained. 28.67 g **28** as a pale beige powder was obtained (yield: 81% based on 99.6% HPLC-purity, considering the additional product). Melting range: 238-243°C. HPLC (method 2): t_R = 3.30 min (99.6%). Chiral HPLC (method 3): 100%ee, t_R (4*S*,5a*R*,8a*R*-enantiomer) = 14.62 min, t_R (4*R*,5a*S*,8a*S*-enantiomer 2) = 25.28 min. LCMS (method 1): t_R = 0.64 min (98.2%, product), m/z 288.3 $[M+H]^+$. ^{1}H NMR (600 MHz, DMSO-d_6) δ 8.01 (s, 1H), 7.48 – 7.44 (m, 2H), 7.43 – 7.35 (m, 3H), 5.01 – 4.89 (m, 2H), 3.83 (d, J = 7.8 Hz, 1H), 3.61 (td, J = 3.7, 1.9 Hz, 1H), 3.27 (dd, J = 9.9, 6.1 Hz, 1H), 2.90 (dddd, J = 11.9, 4.0, 2.7, 1.2 Hz, 1H), 2.83 – 2.73 (m, 1H), 2.64 (d, J = 11.9 Hz, 1H), 2.22 (ddt, J = 14.2, 8.4, 3.0 Hz, 1H), 1.41

(ddd, J = 14.3, 9.2, 1.9 Hz, 1H), 1 proton not visible. LC-HRMS: calcd for $C_{15}H_{18}N_3O_3$ $[M+H]^+$ 288.13427, found 288.13424. X-ray crystal structure data available in SI.

(4*R*,5a*S*,8a*S*)-3-Hydroxyhexahydro-2*H*-1,4-methanopyrrolo[3,4-d][1,3]diazepine-2,8(3*H*)-dione (29). 28 (5 g, 17.4 mmol) was dissolved in methanol/DCM 1:1 (87 mL) and the solution was degassed with nitrogen. Pd/C 10% Degussa type 101 NE/W (50% water) (1.85 g, 0.87 mmol, 0.05 equiv) was added, the atmosphere replaced by hydrogen (balloon) and the reaction mixture stirred for 90 min. The reaction mixture was filtered through celite, eluting with MeOH/DCM:MeOH 1:1 (0.5 L) and concentrated to afford 3.42 g **29** (quant. yield) of a white solid that was used crude in the next step. LCMS (method 3): t_R = 0.13 min; m/z 198 $[M+H]^+$.

Tetrabutylammonium (4*R*,5a*S*,8a*S*)-2,8-dioxohexahydro-2*H*-1,4-methanopyrrolo[3,4-d][1,3]diazepin-3(4*H*)-yl sulfate (30). 29 (3.42 g, 17.3 mmol) was dissolved in pyridine (87 ml), SO_3·pyridine (14.1 g, 87 mmol, 5 equiv) was added and the reaction stirred at rt for 18 h. The reaction mixture was filtered through a disposable plastic filter and concentrated

(bath temp < 30°C). The resulting material was dissolved in sat. NaH_2PO_4 (0.5 L) and washed with EtOAc (0.3 L) in a separation funnel. To the aqueous phase was added tetrabutylammonium hydrogen sulfate (8.83 g, 26.0 mmol, 1.5 equiv) and the mixture was stirred for 30 min at rt and then extracted with DCM (2 × 0.5 L), dried over sodium sulfate, filtered and concentrated. This material was purified by chromatography using a 330 g RediSep silica gel column (DCM load) eluting with 0-30% MeOH in DCM and concentrated. The residue was dissolved in DCM, filtered through a plastic disposable filter and concentrated to afford 7.63 g of **30** as a white foam (yield: 85%). The product was used without further purification. LCMS (method 3): t_R = 0.14 min; m/z 278 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO-d6) δ 7.99 (s, 1H), 3.97 (br s, 1H), 3.81 (d, J=7.8 Hz, 1H), 3.28 (dd, J=6.2, 9.9 Hz, 1H), 3.20 - 3.12 (m, 8H), 2.98 (br d, J=11.9 Hz, 1H), 2.78 (d, J=9.9 Hz, 1H), 2.65 (d, J=11.9 Hz, 1H), 2.47 (s, 1H), 2.27 - 2.18 (m, 1H), 1.56 (quin, J=7.8 Hz, 8H), 1.45 - 1.36 (m, 1H), 1.31 (sxt, J=7.3 Hz, 8H), 0.93 (t, J=7.3 Hz, 12H).

Sodium (4*R*,5a*S*,8a*S*)-2,8-dioxohexahydro-2*H*-1,4-methanopyrrolo[3,4-d][1,3]diazepin-3(4*H*)-yl sulfate (1). To a 250 mL round bottom flask with a magnetic stir bar was added

30 (6.9 g, 13.30 mmol) followed by isobutanol (20.8 mL) and water (1.35 mL) to give a clear solution. Sodium 2-ethylhexanoate (4.56 g, 26.6 mmol, 2 equiv) dissolved in isobutanol (20.8 mL) and water (1.35 mL) was added via syringe pump at 8 mL/h at 40°C. The mixture was then stirred for 1 h at 40°C and then cooled to rt and stirred overnight. The mixture was filtered with a Whatman qualitative filter paper 90 mm cat No 1001090. The cake was washed with n-butanol (3 ×) and then ice cold acetone (3 ×). The cake was dried by sucking nitrogen through it for 3 h and then lyophilized to afford 3.05 g **1** as a white solid (yield: 73% based on an estimated NMR-purity of 95%). The last three steps were repeated two more times to obtain in total 10 g **1**. $[\alpha]_D^{20}$ –9.3 (c=0.5 w/v%, DMSO). HRMS (ESI, Synapt G2 HDMS (Waters), TOF mass spectrometer): calcd for $C_8H_{12}N_3O_6S$ $[M+H]^+$ 278.0447, found 278.0453. Elemental analysis (performed at Robertson Microlit Laboratories): found: C, 31.74; H, 3.15; N, 13.68; S, 8.5; Na, 7.67. Calc. for $C_8H_{10}N_3NaO_6S$: C, 32.11; H, 3.37; N, 14.04; S, 10.72; Na, 7.68. ^{1}H NMR (Bruker AVANCE III-500, 400 MHz, D_2O) δ 4.12 – 4.00 (m, 2H), 3.37 (dd, J = 10.7, 6.2 Hz, 1H), 3.18 (dddd, J = 12.3, 4.2, 2.7, 1.4 Hz, 1H), 2.95 (d, J = 10.7 Hz, 1H), 2.80 (d, J = 12.2 Hz, 1H), 2.66 (qd, J = 8.6, 6.1 Hz, 1H), 2.35 (ddt, J = 14.7, 8.7, 3.0 Hz, 1H), 1.50 (ddd, J = 14.8, 9.1,

1.9 Hz, 1H). ^{13}C NMR (Bruker AVANCE III-500, 125.76 MHz, D_2O) δ 174.35, 168.30, 62.38, 58.7, 47.5, 45.56, 28.47, 26.2.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI:

Crystallographic information, copies of ^1H NMR, chiral HPLC, HRMS, DSC when applicable.

AUTHOR INFORMATION

* To whom correspondence should be addressed. Tel: +41 61 6964576. E-mail: markus.furegati@novartis.com.

ACKNOWLEDGMENT

The authors would like to thank Kurt Laumen and Claude Haby for the enzymatic screen;

Doris Brandenberger, Fabian Bruhin, Ina Dix, Corinne Durand, Dan Huyn, Marie-Anne Lozach, Alexandre Luneau, Corinne Marx, Caroline Radoch, Dominik Rufle, Harald Schröder and Trixie Wagner for technical assistance and analytical support; and Cara Brocklehurst for proof reading.

REFERENCES

1. Reck, F.; Bermingham, A.; Blais, J.; Casarez, A.; Colvin, R.; Dean, C. R.; Furegati, M.; Gamboa, L.; Growcott, E.; Li, C.; Lopez, S.; Metzger, L.; Nocito, S.; Ossola, F.; Phizackerley, K.; Rasper, D.; Shaul, J.; Shen, X.; Simmons, R. L.; Tang, D.; Tashiro, K.; Yue, Q., IID572: A New Potentially Best-In-Class β-Lactamase Inhibitor. *ACS Infectious Diseases* **2019,** *5*, 1045-1051.

2. PROCESSES FOR PREPARING HETEROCYCLIC COMPOUNDS INCLUDING TRANS-7-OXO-6-(SULPHOOXY)-1,6-DIAZABICYCLO[3,2,1]OCTANE-2-CARBOXAMIDE AND SALTS THEREOF. US2012/323010, 2012.

3. Ball, M.; Boyd, A.; Ensor, G. J.; Evans, M.; Golden, M.; Linke, S. R.; Milne, D.; Murphy, R.; Telford, A.; Kalyan, Y.; Lawton, G. R.; Racha, S.; Ronsheim, M.; Zhou, S. H., Development of a Manufacturing Route to Avibactam, a β-Lactamase Inhibitor. *Organic Process Research & Development* **2016,** *20*, 1799-1805.

4. Chu, L.; Ohta, C.; Zuo, Z.; MacMillan, D. W. C., Carboxylic Acids as A Traceless Activation Group for Conjugate Additions: A Three-Step Synthesis of (±)-Pregabalin. *J. Am. Chem. Soc.* **2014,** *136*, 10886-10889.

5. Waldner, A., [4 + 2]-Cycloadditionen von α,β-ungesättigten Hydrazonen. Teil 1. Pyridin-2,3-dicarboximide aus 1-(Dimethylamino)-1,4-dihydropyridin-Derivaten. *Helvetica Chimica Acta* **1988,** *71*, 486-492.

6. Van, C. T.; Zdobinsky, T.; Seebohm, G.; Nennstiel, D.; Zerbe, O.; Scherkenbeck, J., Structural Prerequisites for Receptor Binding of Helicokinin I, a Diuretic Insect Neuropeptide from Helicoverpa zea. *European Journal of Organic Chemistry* **2014,** *2014*, 2714-2725.

7. Christie, B. D.; Rapoport, H., Synthesis of optically pure pipecolates from L-asparagine. Application to the total synthesis of (+)-apovincamine through amino acid decarbonylation and iminium ion cyclization. *The Journal of Organic Chemistry* **1985,** *50*, 1239-1246.

8. Zhou, H.; Sun, G.; Liu, Z.; Zhan, X.; Mao, Z., Synthesis of 4-methoxy and 5-methoxy substituted 7-aza-isoindolin-1-ones. *Heterocycles* **2013,** *87*, 2071 - 2079.

9. Wang, T.; Du, L.-D.; Wan, D.-j.; Li, X.; Chen, X.-Z.; Wu, G.-F., Use of Lipase Catalytic Resolution in the Preparation of Ethyl (2S,5R)-5-((Benzyloxy)amino)piperidine-2-carboxylate, a Key Intermediate of the β-Lactamase Inhibitor Avibactam. *Organic Process Research & Development* **2018,** *22*, 1738-1744.

10. Alvaro, G.; Andreotti, D.; Bergauer, M.; Giovannini, R.; Marasco, A. Prolinamide derivatives as sodium channel modulators. WO2007042250A1, 2007.

11. Charton, J.; Gauriot, M.; Totobenazara, J.; Hennuyer, N.; Dumont, J.; Bosc, D.; Marechal, X.; Elbakali, J.; Herledan, A.; Wen, X.; Ronco, C.; Gras-Masse, H.; Heninot, A.; Pottiez, V.; Landry, V.; Staels, B.; Liang, W. G.; Leroux, F.; Tang, W.-J.; Deprez, B.; Deprez-Poulain, R., Structure-activity relationships of imidazole-derived 2-[N-carbamoylmethyl-alkylamino]acetic acids, dual binders of human insulin-degrading enzyme. *European Journal of Medicinal Chemistry* **2015,** *90*, 547 - 567.

12. Marx, M. A.; Grillot, A.-L.; Louer, C. T.; Beaver, K. A.; Bartlett, P. A., Synthetic Design for Combinatorial Chemistry. Solution and Polymer-Supported Synthesis of

Polycyclic Lactams by Intramolecular Cyclization of Azomethine Ylides. *Journal of the American Chemical Society* **1997,** *119*, 6153-6167.

13. A 48 g trial batch was purified on 1.2 kg silica gel in three portions, the product fraction still contained an unacceptable level of isomer impurity. However, we found that this diastereoisomer was depleted along the synthesis in particular after the crystallization of compound **21**. The large batch was therefore used crude.

14. $LiBH_4$, although slow, gave the best yield at rt (heating resulted in the reduction of the lactam). $NaBH_4$/LiCl in EtOH gave only 50% isolated yield, the reaction mixture was a suspension followed by a messy workup.

15. Mena, M.; Bonjoch, J.; Pardo, D. G.; Cossy, J., Ring expansion of functionalized octahydroindoles to enantiopure cis-decahydroquinolines. *Journal of Organic Chemistry* **2006,** *71*, 5930 - 5935.

16. Jarvis, S. B. D.; Charette, A. B., Synthesis of Enantiopure Substituted Piperidines via an Aziridinium Ring Expansion. *Organic Letters* **2011,** *13*, 3830-3833.

17. Proper sample work up for reaction monitoring was important: quenching of an aliquot of the reaction mixture with aq. NaOH prior to analysis stopped the equilibration and gave meaningful data, whereas simple quench in ACN did not provide reliable isomer ratio data.

18. Groth, U.; Richter, L.; Schoellkopf, U., Synthesis of Substituted Dibenzo<a,d>azepines via a Base-Mediated Ring Expansion. *Liebigs Annalen der Chemie* **1992**, 199 - 202.

19. Mitsunobu, O., The Use of Diethyl Azodicarboxylate and Triphenylphosphine in Synthesis and Transformation of Natural Products. *Synthesis* **1981,** *1981*, 1-28.

20. Yamashita, T.; Kawai, N.; Tokuyama, H.; Fukuyama, T., Stereocontrolled total synthesis of (-)-eudistomin C. *Journal of the American Chemical Society* **2005,** *127*, 15038 - 15039.

21. A method for synthesizing intermediate avey Batan (by machine translation). CN106831772, 2017.

22. Reddy, P. A.; Schall, O. F.; Wheatley, J. R.; Rosik, L. O.; McClurg, J. P.; Marshall, G. R.; Slomczynska, U., O-Protected N-(2-

Nitrophenylsulfonyl)hydroxylamines: Novel Reagents for the Synthesis of Hydroxamates. *Synthesis* **2001**, 1086 - 1092.