Atroposelective Total Synthesis of Darobactin A

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Supporting Information Placeholder

ABSTRACT: A concise, modular synthesis of the novel antibiotic darobactin A is disclosed. The synthesis successfully forges the hallmark strained macrocyclic ring systems in a sequential fashion. Key transformations include two atroposelective Larock-macrocylizations, one of which proceeds with exquisite regioselectivity despite bearing an unprotected alkyne. The synthesis is designed with medicinal chemistry considerations in mind, appending key portions of the molecule at a late-stage. Requisite unnatural amino acid building blocks are easily prepared in enantiopure form using C–H activation and decarboxylative cross-coupling tactics.

Darobactin A (**1**, Figure 1), isolated in 2019 by the Lewis group from *Photorhabdus* bacteria[1], features a highly strained fused macrocyclic peptide-based ring system. It exhibits selective antibiotic activity against Gram-negative pathogens in both in vitro and animal infection models and functions through an intriguing new mode of action. The rigid structure of darobactin A is likely responsible for its activity, based on crystallographic studies.[2] The fact that all natural analogs retain the unique core skeleton[3-4], which acts as a β-strand mimetic[2], supports this notion. Architecturally ornate peptide-based macrocycles have a rich history of acting as promising antibacterial agents from the clinically validated vancomycins[5-8] to the more recently pursued arylomycins.[9-11] Darobactin A contains multiple bonds that have not been previously described in antibiotic natural products[12-13]–namely, an aliphatic-aromatic ether moiety between the two tryptophan residues and a C-C bond between the β-carbon of lysine and C6 of the central tryptophan indole. Presumably, these bonds are formed in Nature via post-translational macrocyclizations of a linear peptide.[1,14] These unusual features form the basis of a fused macrocylic bicycle (14- and 15-membered rings) containing atropisomeric indoles.[15-16] A fully synthetic route to darobactin A would not only increase the available supply of material for further study, but also provide a framework for medicinal chemistry explorations.[2,17] In this Communication, a modular total synthesis of **1** that effectively addresses its structural and stereochemical challenges is disclosed.[18]

Although the final retrosynthetic blueprint employed to arrive at **1** is depicted in Figure 1, it is the culmination of multiple failed routes that preceeded it. Strategies wedded to macrolactamization, Suzuki, and Heck tactics were all extensively investigated.[A] Ultimately an approach featuring Larock-based macrocylizations[15,19-21] successfully accessed the natural product using inexpensive amino acids and arenes as building blocks (Figure 1, **2**-**8**).

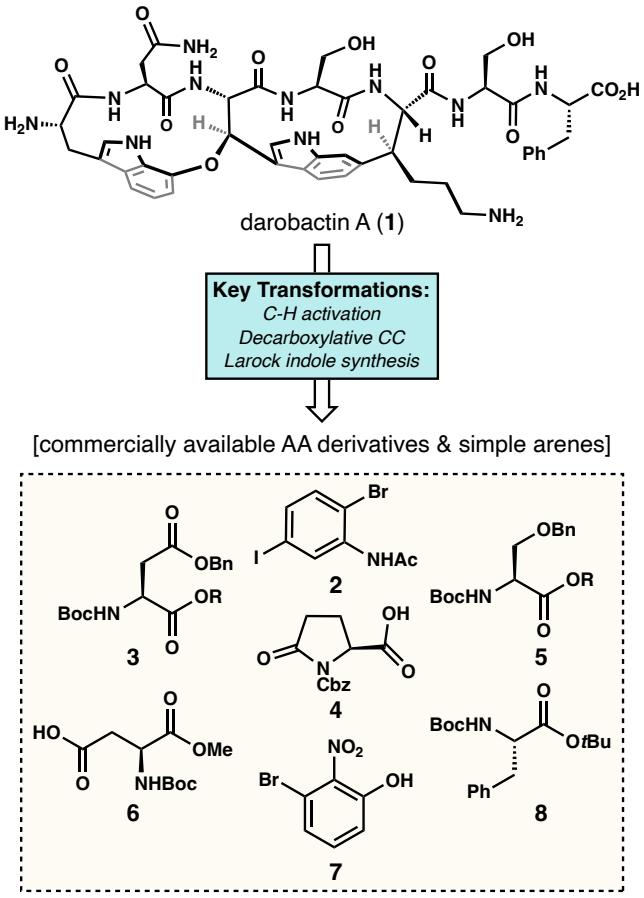


Figure 1. Darobactin A (**1**): Retrosynthetic strategies and building blocks.

To begin, two of the key unnatural amino acids required for the synthesis were fashioned using decarboxylative cross coupling methodology (Scheme 1A). Previously, such derivatives were prepared from serine[22] in two steps and required the use of copper cyanide[23,24] or from 2-prop-2-ynoxyoxane in four steps[21,25] using Schöllkopf's chiral auxiliary.[26] By employing a decarboxylative alkynylation approach[27], a scalable synthesis of **9** was achieved in one step from commercially available **6**. Alkyne-dipeptides **12** and **14** could be divergently accessed from **6,** either through SeO_2-based

oxidation (via **10**/**11**) followed by peptide coupling or through peptide coupling (via **13**) and deprotection, respectively. Although diastereomer **11** ultimately proved to be the desired isomer, both **10** and **11** were needed in this study (*vide infra*).

Construction of the core of **1** (Scheme 1B) began with formation of the C-C bond between the lysine and tryptophan moieties. Employing C-H functionalization logic[28] to access this unique connectivity, an 8-aminoquinoline(AQ)-directed[29-30] palladium-catalyzed, stereospecific C-H arylation[31] was performed using conditions developed by Schreiber and co-workers[32] to couple pyroglutamate derivative **15** (synthesized from **3** and 8-aminoquinoline, see SI) and arene **2** in a *syn* fashion on multi-gram scale to give **16** (51% yield). Reductive ring opening[33] of **16** followed by benzoyl protection of the resulting primary alcohol provided intermediate **17** (69% yield). Following a Cbz deprotection[34] **12** was appended in the presence of HATU (53% yield) to set the stage for testing the Larock cyclization.

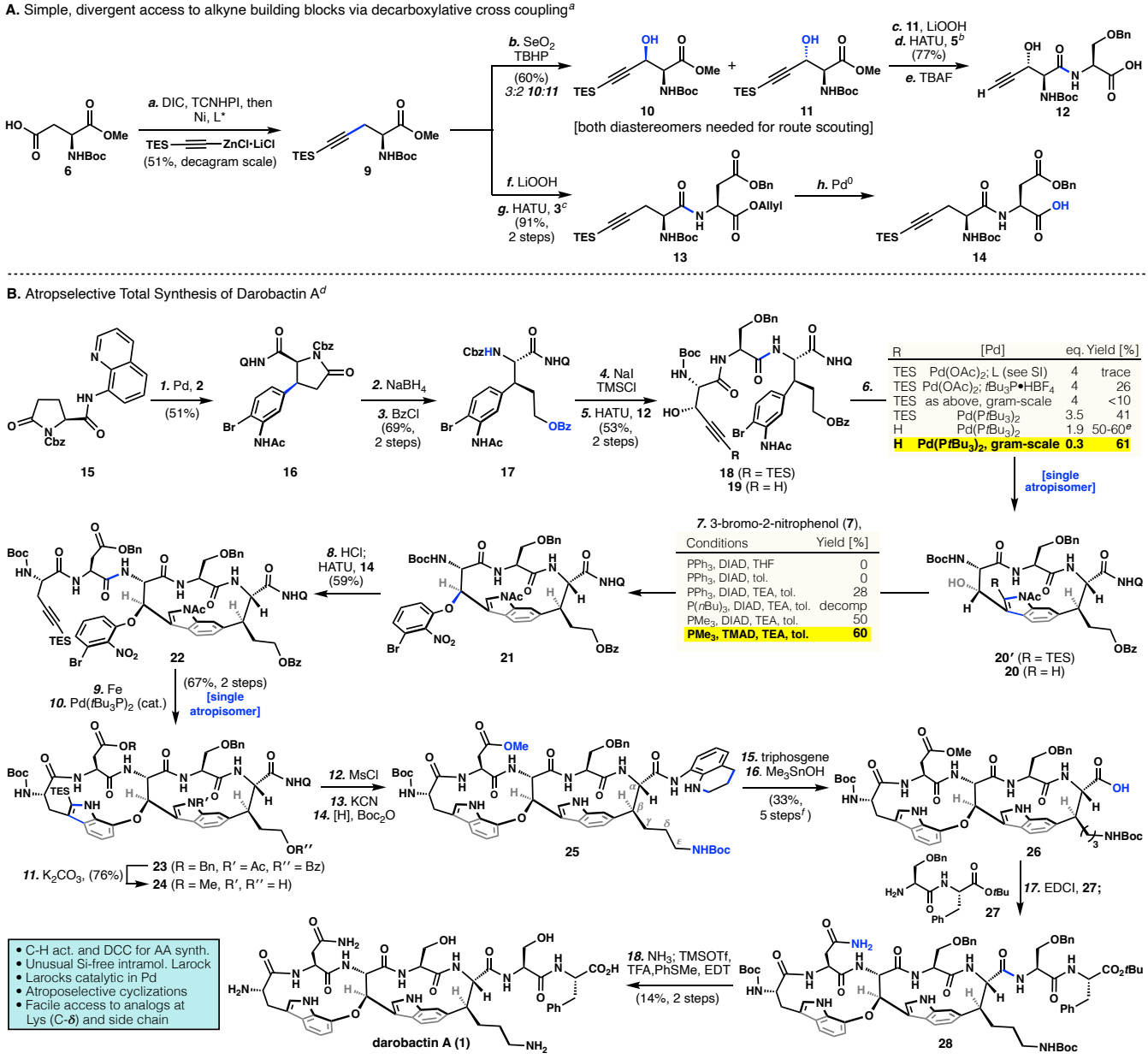
Over a year of research went into developing conditions for this reaction and the related cyclization derived from alkyne **10**. Early development was performed on **18** and suffered from the use of superstoichiometric Pd, poor scalability, deleterious dehalogenation, atropisomer mixtures, and acyl transfer-derived byproducts (from indole N to hydroxy group). As depicted in the inset table (step 6), multiple ligands were evaluated with tBu$_3$P•HBF$_4$ emerging as the most effective. Using 4 equivalents of Pd, a 26% yield of macrocycle **20´** was obtained on mg scale. Unfortunately, this reaction suffered from scalability issues above 100 mg. In accord with studies from the Reisman[24] and Boger[21] labs, pre-formed Pd(PtBu$_3$)$_2$ provided a glimmer of hope by allowing a slight reduction in Pd-loading (3.5 equivalents) and delivering 41% yield of **20´** along with a significant amount of the des-bromo byproduct. Since oxidative addition on **18** must be facile (as judged by extensive debromination), a combination of unfavorable steric interactions and strain likely hinder this cyclization. Traditionally, the Larock indole synthesis requires a hindered alkyl or silyl-terminated alkyne to provide the desired regiochemical outcome.[35] It was reasoned that in this specific context, a triethylsilyl (TES) blocking group on **18** may actually be superfluous as the regiochemistry may be controlled by the conformation of the cyclization precursor. Gratifyingly, unprotected alkyne **19** engaged smoothly in Larock macrocyclization to deliver **20** in up to 60% yield, albeit using 1.9 equiv of Pd. Further optimization efforts were directed towards reducing the Pd loading to substoichiometric levels. Interstingly, the Pd loading could be reduced to 30 mol%, rendering the reaction catalytic, without detrimental impact to the yield. Ultimately the macrocyclization could be efficiently conducted on a gram-scale, furnishing **20** as a single atropisomer in 61% isolated yield. A small amount of byproducts with the same mass yet different polarity can be observed on LC, however appreciable amounts could not be isolated for characterization.

Given the success of the Larock reaction in forging the central ring, this strategy was also pursued as a means to perform the second macrocyclization. This disconnection required installation of an ortho bromoaniline functionality onto the hydroxyl group of **20** with inversion of the stereochemistry. A variety of etherification tactics were evaluated using alkynes derived from both **10** and **11** in order to test both stereoretentive (i.e. S$_N$Ar) and stereoinvertive strategies. Ultimately, Mitsunobu displacement of the alcohol with 3-bromo-2-nitrophenol (**7**) proved to be the most successful. Through extensive reagent and solvent screening (see inset table, step 7), it was revealed that the unusual combination of PMe$_3$ and TMAD[36-40] in toluene afforded **21** in 60% isolated yield (500 mg scale). Historically such reagents have been employed for carbon-based nucleophiles; in this case the use of these conditions led to increased reactivity with considerable suppression of two major reaction byproducts (β-elimination and intramolecular cyclization of the N-Boc group to an oxazolidinone). With the key bromo arene installed with the proper stereochemistry, Boc-deprotection of **21** and subsequent coupling with **14** using HATU afforded **22** (59% yield). Reduction of the nitro group of compound **22** to the primary amine was completed using Fe and NH$_4$Cl.[41] Gratifyingly, the Larock cyclization of the corresponding crude aniline was successful using similar conditions to the first macrocyclization reaction to give **23** as a single atropisomer in 67% isolated yield over two steps.

Inherent to the design of this synthesis was a desire to probe the SAR of the lysine residue as crystallographic studies indicate the importance of this side chain as indicated by its interactions with the phosphate moieties of lipids.[2] The ability to access a library of analogs requires late-stage homologation to arrive at **1**. Thus, installation of a cyano group followed by reduction provided the desired side chain through a simple four step sequence: (a) methanolysis of **23** to **24** (76% yield), (b) mesylation and concomitant TES deprotection in the presence of methanesulfonic acid, (c) cyanation under phase transfer conditions, and (d) reduction of both the nitrile and AQ ring with in situ generated nickel boride[42] followed by in situ Boc protection. The fortuitous reduction of the AQ allowed for its smooth removal[43] using conditions developed by Geyer[44] (as inspired by methodology from the Dawson group).[45] Consequently, treatment of **25** with triphosgene followed by hydrolysis with trimethyltin hydroxide[46] delivered **26** in 33% yield over 5 steps. With the carboxylic acid revealed, conditions were evaluated to install the side chain **27** (synthesized from **5** and **8**, see SI). Numerous peptide coupling reagents were screened for this transformation, however successful amide bond formation was only observed in appreciable quantities using a combination of EDCI and HOAt.[47] The product of the coupling reaction was then subjected to ammonolysis to convert the aspartate residue to the asparagine **28**. At this stage, all that remained was global deprotection, which was realized by treatment of **28** with a combination of TMSOTf, TFA, thioanisole, and ethane dithiol[48] to complete the synthesis of **1** in 14% yield for the final sequence (18 steps LLS from **15**).

In addition to exquisite control of core stereochemical features, this synthesis highlights the unique ability of Larock cyclization to generate two strained ring systems. In one of those instances specific geometrical and conformational constraints guide regiochemistry; this allows for the use of an unprotected alkyne, rendering it catalytic in Pd, and providing a single atropisomeric outcome. At this juncture a definitive rationalization for the observed atroposelectivity is not apparent. The precise choreography of cyclization events on specific substrates were planned using plastic molecular models which supported the observed outcomes. Rapid access to key unnatural AA building blocks from inexpensive natural AAs is enabled by scalable C–H activation and decarboxylative coupling methods. Although non-strategic from an efficiency standpoint, homologation is deliberately enlisted late-stage to install the lysine sidechain solely for medicinal chemistry explorations of this portion of the molecule. For the same reasons, a late-stage incorporation of the C-terminus dipeptide was pursued. The atroposelective and modular synthesis of **1** sets the stage for the generation of analogs that are currently unavailable through engineered biosynthetic approaches and enables future exploration of the potent antibiotic activity of this important family of peptidic natural products.

Scheme 1. Total synthesis of darobactin A



[a]Reagents and conditions, alkyne fragment syntheses: (***a***) $NiCl_2$•$6H_2O$ (50 mol%), L* = *t*Bubpy (50 mol%), TES-alkynyl-ZnCl•LiCl (0.2M in THF, 3.5 equiv, see SI for preparation.), DMF, rt, 4 h. (***b***) SeO_2 (2.00 equiv), TBHP (4.00 equiv), MeCN, 50 °C, 4 h. (***c***) LiOH (1M aqueous, 1.50 equiv), H_2O_2 (30 wt.% aqueous, 15.0 equiv), THF, 0 °C to rt, 4 h. (***d***) **11**, hydrolyzed (1.10 equiv), **5** (1.00 equiv), HATU (1.30 equiv), DIPEA (3.30 equiv). [b]**5** (R = SEM), Boc-deprotected prior to coupling. (***e***) TBAF (3.00 equiv), THF, 0 °C, 1 h. (***f***) LiOH (1M aqueous, 1.50 equiv), H_2O_2 (30 wt.% aqueous, 15.0 equiv), THF, 0 °C to rt, 4 h. (***g***) **9**, hydrolyzed (1.90 equiv), **3** (1.00 equiv), HATU (1.90 equiv), DIPEA (10.0 equiv). [c]**3** (R = allyl), Boc-deprotected prior to coupling. (***h***) $Pd(PPh_3)_4$ (0.10 equiv), morpholine (5.00 equiv), THF, rt, 20 min. [d]Reagents and conditions, darobactin A synthesis: (***1***) **2** (1.00 equiv), **15** (1.80 equiv), AgOAc (1.50 equiv), dibenzyl hydrogen phosphate (0.20 equiv), $Pd(OAc)_2$ (60 mol%), CMPE, 120 °C, 16 h. (***2***) $NaBH_4$ (4.00 equiv), 8:1 THF:*t*BuOH, rt, 2 h. (***3***) benzoyl chloride (8.0 equiv), pyridine (12.0 equiv), DMAP (0.10 equiv), DCM, rt, 4 h. (***4***) NaI (25.0 equiv), TMSCl (15.0 equiv), rt, 3 h. (***5***) HCl (4M in dioxane); **12** (1.20 equiv), HATU (1.20 equiv), DIPEA (3.00 equiv). (***6***) $Pd(PtBu_3)_2$ (30 mol%), DIPEA (5.00 equiv), dioxane, 80 °C, 1.5 h. (***7***) 3-bromo-2-nitrophenol **7** (20.0 equiv), TEA (10.0 equiv), PMe_3 (1M in toluene, 10.0 equiv), PMe_3 (1M in THF, 3.0 equiv), TMAD (13.0 equiv), 38 °C, rt, 48 h. (***8***) HCl (4M in dioxane); **14** (1.75 equiv), HATU (1.75 equiv), DIPEA (8.00 equiv). (***9***) Fe powder (120 equiv), NH_4Cl (40.0 equiv), AcOH, 50 °C, 4 h. (***10***) $Pd(PtBu_3)_2$ (50 mol%), DIPEA (5.00 equiv), dioxane, 80 °C, 2 h. (***11***) K_2CO_3 (16.0 equiv), MeOH, rt, 6 h. (***12***) TEA (19.0 equiv), MsCl (35.0 equiv), DCM, 0 °C, 1 h.; MsOH (1.10 equiv), DCM, rt, 12 h. (***13***) KCN (100 equiv), TBHS (7.00 equiv), KH_2PO_4 (39.0 equiv), 1:1 H_2O:$CHCl_3$, 36 °C, 48 h. (***14***) $NiCl_2$•$6H_2O$ (3.00 equiv), Boc_2O (15.0 equiv), $NaBH_4$ (16.0 equiv), 3:1 MeOH:THF, -10 °C, 3 h. (***15***) DIPEA (9.00 equiv), triphosgene (2.90 equiv), DCM, 0 °C, 5 h. (***16***) Me_3SnOH (13.0 equiv), DCE, 31 °C, 6 h. (***17***) **27** (8.00 equiv), EDCI (4.00 equiv), HOAt (4.00 equiv), $NaHCO_3$ (6.00 equiv), DMF:DCM 1:1, 33 °C, 4 h. (***18***) NH_3(*g*) sparge with balloon, MeOH, 33 °C, 48 h; TFA:TMSOTf:thioanisole:EDT 12:3:2:1 0 °C, 2 h. [e]LCMS yield. [f]Intermediates at steps 13 and 14 were column purified but contained inseparable contaminants derived from reaction conditions (i.e. tetrabutylammonium). Semi-pure fractions were carried forward

but an accurate yield could not be determined at these steps. Abbreviations: DIC = *N,N*-diisopropylcarbodiimide; TCNHPI = tetrachloro-*N*-hydroxy phthalimide; *t*Bubpy = 4,4'-ditertbutyl-2,2'-bipyridine; TES = triethylsilyl; TBHP = tertbutylhydroperoxide; THF = tetrahydrofuran; SEM = 2-(trimethylsilyl)ethoxymethyl; HATU = (1-[bis(dimethylamino)methylene]-1H-1,2,3-triazolo[4,5-b]pyridinium 3-oxide hexafluorophosphate; BzCl = benzoyl chloride; Q = 8-aminoquinoline; DCM = dichloromethane; TMSCl = trimethylsilyl chloride; DIAD = diisopropyl azodicarboxylate; tol. = toluene; TMAD = tetramethylazodicarboximide; MsCl = methanesulfonyl chloride; MsOH = methanesulfonic acid; TBHS = tetrabutylammonium hydrogensulfate; EDCI = 1-ethyl-3-(3-methylaminopropyl)carbodiimide hydrochloride; TMSOTf = trimethylsilyl triflate; TFA = trifluoroacetic acid; EDT = ethane dithiol; DMF = *N,N*-dimethylformamide; CPME = cylopentylmethyl ether; DMAP = *N,N*-dimethyl-4-aminopyridine; DIPEA = diisopropylethylamine; TEA = triethylamine; DCE = dichloroethane; HOAt = 1-hydroxy-7-azabenzotriazole.

ASSOCIATED CONTENT

Supporting Information. Experimental procedures, analytical data (^{1}H and ^{13}C NMR, MS) for all new compounds as well as optimization tables. This material is available free of charge *via* the Internet at http://pubs.acs.org.

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Author Contributions

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Notes

The authors declare no competing financial interest.

[A]The details of these failed approaches will be disclosed in a full account of this work. The entirety of those studies as well as an earlier, near complete variant of the current work was disclosed in detail to specific employees of Merck Inc. on June 3, 2021 under a confidentiality agreement.

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REFERENCES

(1) Imai, Y.; Meyer, K. J.; Iinishi, A.; Favre-Godal, Q.; Green, R.; Manuse, S.; Caboni, M.; Mori, M.; Niles, S.; Ghiglieri, M.; Honrao, C.; Ma, X. Y.; Guo, J. J.; Makriyannis, A.; Linares-Otoya, L.; Boehringer, N.; Wuisan, Z. G.; Kaur, H.; Wu, R.; Mateus, A.; Typas, A.; Savitski, M. M.; Espinoza, J. L.; O'Rourke, A.; Nelson, K. E.; Hiller, S.; Noinaj, N.; Schäberle, T. F.; D'Onofrio, A.; Lewis, K., A new antibiotic selectively kills Gram-negative pathogens. *Nature* **2019,** *576,* 459–464.

(2) Kaur, H.; Jakob, R. P.; Marzinek, J. K.; Green, R.; Imai, Y.; Bolla, J. R.; Agustoni, E.; Robinson, C. V.; Bond, P. J.; Lewis, K.; Maier, T.; Hiller, S., The antibiotic darobactin mimics a beta-strand to inhibit outer membrane insertase. *Nature* **2021,** *593,* 125–129.

(3) Wuisan, Z. G.; Kresna, I. D. M.; Bohringer, N.; Lewis, K.; Schäberle, T. F., Optimization of heterologous Darobactin A expression and identification of the minimal biosynthetic gene cluster. *Metab. Eng.* **2021,** *66,* 123–136.

(4) Bohringer, N.; Green, R.; Liu, Y.; Mettal, U.; Marner, M.; Modaresi, S. M.; Jakob, R. P.; Wuisan, Z. G.; Maier, T.; Iinishi, A.; Hiller, S.; Lewis, K.; Schäberle, T. F., Mutasynthetic Production and Antimicrobial Characterization of Darobactin Analogs. *Microbiol. Spectr.* **2021,** *9,* e01535-21.

(5) Bruniera, F. R.; Ferreira, F. M.; Saviolli, L. R. M.; Bacci, M. R.; Feder, D.; Pedreira, M. D. G.; Peterlini, M. A. S.; Azzalis, L. A.; Junqueira, V. B. C.; Fonseca, F. L. A., The use of vancomycin with its therapeutic and adverse effects: a review. *Eur. Rev. Med. Pharmaco.* **2015,** *19,* 694–700.

(6) Okano, A.; Isley, N. A.; Boger, D. L., Total Syntheses of Vancomycin-Related Glycopeptide Antibiotics and Key Analogues. *Chem. Rev.* **2017,** *117,* 11952–11993.

(7) Okano, A.; Isley, N. A.; Boger, D. L., Peripheral modifications of [Psi[CH2NH]Tpg(4)]vancomycin with added synergistic mechanisms of action provide durable and potent antibiotics. *Proc. Natl. Acad. Sci. U.S.A.* **2017,** *114,* E5052–E5061.

(8) Moore, M. J.; Qu, S. W.; Tan, C. H.; Cai, Y.; Mogi, Y.; Keith, D. J.; Boger, D. L., Next-Generation Total Synthesis of Vancomycin. *J. Am. Chem. Soc.* **2020,** *142,* 16039–16050.

(9) Liu, J.; Smith, P. A.; Steed, D. B.; Romesberg, F., Efforts toward broadening the spectrum of arylomycin antibiotic activity. *Bioorg. Med. Chem. Lett.* **2013,** *23,* 5654–5659.

(10) Smith, P. A.; Koehler, M. F. T.; Girgis, H. S.; Yan, D.; Chen, Y.; Chen, Y.; Crawford, J. J.; Durk, M. R.; Higuchi, R. I.; Kang, J.; Murray, J.; Paraselli, P.; Park, S.; Phung, W.; Quinn, J. G.; Roberts, T. C.; Rouge, L.; Schwarz, J. B.; Skippington, E.; Wai, J.; Xu, M.; Yu, Z.; Zhang, H.; Tan, M. W.; Heise, C. E., Optimized arylomycins are a new class of Gram-negative antibiotics. *Nature* **2018,** *561,* 189–194.

(11) Peters, D. S.; Romesberg, F. E.; Baran, P. S., Scalable Access to Arylomycins via C-H Functionalization Logic. *J. Am. Chem. Soc.* **2018,** *140,* 2072–2075.

(12) Swain, J. A.; Walker, S. R.; Calvert, M. B.; Brimble, M. A., The tryptophan connection: cyclic peptide natural products linked via the tryptophan side chain. *Nat. Prod. Rep.* **2022,** *39,* 410–443.

(13) Crich, D.; Banerjee, A., Expedient synthesis of threo-beta-hydroxy-alpha-amino acid derivatives: phenylalanine, tyrosine, histidine, and tryptophan. *J. Org. Chem.* **2006,** *71,* 7106–7109.
(14) Guo, S.; Wang, S.; Ma, S. Z.; Deng, Z. X.; Ding, W.; Zhang, Q., Radical SAM-dependent ether crosslink in daropeptide biosynthesis. *Nat. Commun.* **2022,** *13,* 2361.
(15) Garfunkle, J.; Kimball, F. S.; Trzupek, J. D.; Takizawa, S.; Shimamura, H.; Tomishima, M.; Boger, D. L., Total Synthesis of Chloropeptin II (Complestatin) and Chloropeptin I. *J. Am. Chem. Soc.* **2009,** *131,* 16036–16038.
(16) Reisberg, S. H.; Gao, Y.; Walker, A. S.; Helfrich, E. J. N.; Clardy, J.; Baran, P. S., Total synthesis reveals atypical atropisomerism in a small-molecule natural product, tryptorubin A. *Science* **2020,** *367,* 458–463.
(17) Gross, S.; Panter, F.; Pogorevc, D.; Seyfert, C. E.; Deckarm, S.; Bader, C. D.; Herrmann, J.; Muller, R., Improved broad-spectrum antibiotics against Gram-negative pathogens via darobactin biosynthetic pathway engineering. *Chem. Sci.* **2021,** *12,* 11882–11893.
(18) While this manuscript was being written a total synthesis was disclosed by the Merck-Sarlah team: Nesic, M. R., D. B.; Maturano, J.; Shevlin, M.; Pollack, S. R.; Gauthier Jr., D. R.; Trigo-Mouriño, P.; Zhang, L.-K.; Schultz, D.; McCabe Dunn, J. M.; Campeau, L.-C.; Patel, N. R.; Petrone, D. A.; Sarlah, D., Total Synthesis of Darobactin A. *ChemRxiv* **2022,** 10.26434/chemrxiv-2022-c4mz6.
(19) Shimamura, H.; Breazzano, S. P.; Garfunkle, J.; Kimball, F. S.; Trzupek, J. D.; Boger, D. L., Total Synthesis of Complestatin: Development of a Pd(0)-Mediated Indole Annulation for Macrocyclization. *J. Am. Chem. Soc.* **2010,** *132,* 7776-7783.
(20) Breazzano, S. P.; Poudel, Y. B.; Boger, D. L., A Pd(0)-Mediated Indole (Macro)cyclization Reaction. *J. Am. Chem. Soc.* **2013,** *135,* 1600-1606.
(21) Isley, N. A.; Endo, Y.; Wu, Z. C.; Covington, B. C.; Bushin, L. B.; Seyedsayamdost, M. R.; Boger, D. L., Total Synthesis and Stereochemical Assignment of Streptide. *J. Am. Chem. Soc.* **2019,** *141,* 17361–17369.
(22) Trost, B. M.; Rudd, M. T., Chemoselectivity of the ruthenium-catalyzed hydrative diyne cyclization: Total synthesis of (+)-cylindricine C, D, and E. *Org. Lett.* **2003,** *5,* 4599–4602.
(23) Newhouse, T.; Lewis, C. A.; Baran, P. S., Enantiospecific Total Syntheses of Kapakahines B and F. *J. Am. Chem. Soc.* **2009,** *131,* 6360–6361.
(24) Chuang, K. V.; Kieffer, M. E.; Reisman, S. E., A Mild and General Larock Indolization Protocol for the Preparation of Unnatural Tryptophans. *Org. Lett.* **2016,** *18,* 4750–4753.
(25) Noichl, B. P.; Durkin, P. M.; Budisa, N., Toward Intrinsically Colored Peptides: Synthesis and Investigation of the Spectral Properties of Methylated Azatryptophans in Tryptophan-Cage Mutants. *Biopolymers* **2015,** *104,* 585–600.
(26) Schöllkopf, U.; Groth, U.; Deng, C., Asymmetric Syntheses Via Heterocyclic Intermediates .6. Enantioselective Synthesis of (R)-Amino Acids Using L-Valine as Chiral Agent. *Angew. Chem., Int. Ed.* **1981,** *20,* 798–799.
(27) Smith, J. M.; Qin, T.; Merchant, R. R.; Edwards, J. T.; Malins, L. R.; Liu, Z. Q.; Che, G. D.; Shen, Z. C.; Shaw, S. A.; Eastgate, M. D.; Baran, P. S., Decarboxylative Alkynylation. *Angew. Chem., Int. Ed.* **2017,** *56,* 11906–11910.
(28) Gutekunst, W. R.; Baran, P. S., C-H functionalization logic in total synthesis. *Chem. Soc. Rev.* **2011,** *40,* 1976–1991.
(29) Reddy, B. V. S.; Reddy, L. R.; Corey, E. J., Novel acetoxylation and C-C coupling reactions at unactivated positions in alpha-amino acid derivatives. *Org. Lett.* **2006,** *8,* 3391–3394.
(30) Zaitsev, V. G.; Shabashov, D.; Daugulis, O., Highly regioselective arylation of sp^3 C-H bonds catalyzed by palladium acetate. *J. Am. Chem. Soc.* **2005,** *127,* 13154–13155.
(31) Feng, Y. Q.; Chen, G., Total Synthesis of Celogentin C by Stereoselective C-H Activation. *Angew. Chem., Int. Ed.* **2010,** *49,* 958–961.
(32) Verho, O.; Maetani, M.; Melillo, B.; Zoller, J.; Schreiber, S. L., Stereospecific Palladium-Catalyzed C-H Arylation of Pyroglutamic Acid Derivatives at the C3 Position Enabled by 8-Aminoquinoline as a Directing Group. *Org. Lett.* **2017,** *19,* 4424–4427.
(33) Soai, K.; Oyamada, H.; Takase, M., The Preparation of N-Protected Amino-Alcohols and N-Protected Peptide Alcohol by Reduction of the Corresponding Esters with Sodium-Borohydride - an Improved Procedure Involving a Slow Addition of a Small Amount of Methanol. *Bull. Chem. Soc. Jpn.* **1984,** *57,* 2327–2328.
(34) Olah, G. A.; Narang, S. C.; Gupta, B. G. B.; Malhotra, R., Synthetic Methods and Reactions .62. Transformations with Chlorotrimethylsilane Sodium Iodide, a Convenient in Situ Iodotrimethylsilane Reagent. *J. Org. Chem.* **1979,** *44,* 1247–1251.
(35) Larock, R. C.; Yum, E. K., Synthesis of Indoles Via Palladium-Catalyzed Heteroannulation of Internal Alkynes. *J. Am. Chem. Soc.* **1991,** *113,* 6689–6690.
(36) But, T. Y. S.; Toy, P. H., The mitsunobu reaction: Origin, mechanism, improvements, and applications. *Chem.–Asian J.* **2007,** *2,* 1340–1355.
(37) Swamy, K. C.; Kumar, N. N.; Balaraman, E.; Kumar, K. V., Mitsunobu and related reactions: advances and applications. *Chem. Rev.* **2009,** *109,* 2551–2651.
(38) Reynolds, A. J.; Kassiou, M., Recent Advances in the Mitsunobu Reaction: Modifications and Applications to Biologically Active Molecules. *Curr. Org. Chem.* **2009,** *13,* 1610–1632.
(39) Fletcher, S., The Mitsunobu reaction in the 21st century. *Org. Chem. Front.* **2015,** *2,* 739–752.
(40) Panday, S. K., Advances in the Mitsunobu Reaction: An Excellent Organic Protocol with Versatile Applications. *Mini-Rev. Org. Chem.* **2019,** *16,* 127–140.
(41) Ramadas, K.; Srinivasan, N., Iron-Ammonium Chloride - a Convenient and Inexpensive Reductant. *Synth. Commun.* **1992,** *22,* 3189–3195.
(42) Caddick, S.; Judd, D. B.; Lewis, A. K. D.; Reich, M. T.; Williams, M. R. V., A generic approach for the catalytic reduction of nitriles. *Tetrahedron* **2003,** *59,* 5417–5423.
(43) Fitzgerald, L. S.; O'Duill, M. L., A Guide to Directing Group Removal: 8-Aminoquinoline. *Chem. Eur. J.* **2021,** *27,* 8411–8436.
(44) Nicke, L.; Horx, P.; Harms, K.; Geyer, A., Directed C(sp(3))-H arylation of tryptophan: transformation of the directing group into an activated amide. *Chem. Sci.* **2019,** *10,* 8634–8641.
(45) Blanco-Canosa, J. B.; Nardone, B.; Albericio, F.; Dawson, P. E., Chemical Protein Synthesis Using a Second-Generation N-Acylurea Linker for the Preparation of Peptide-Thioester Precursors. *J. Am. Chem. Soc.* **2015,** *137,* 7197–7209.
(46) Nicolaou, K. C.; Estrada, A. A.; Zak, M.; Lee, S. H.; Safina, B. S., A mild and selective method for the hydrolysis of esters with trimethyltin hydroxide. *Angew. Chem., Int. Ed.* **2005,** *44,* 1378–1382.
(47) Jiang, W. L.; Wanner, J.; Lee, R. J.; Bounaud, P. Y.; Boger, D. L., Total synthesis of the ramoplanin A2 and ramoplanose aglycon. *J. Am. Chem. Soc.* **2002,** *124,* 5288–5290.
(48) Yajima, H.; Fujii, N.; Funakoshi, S.; Watanabe, T.; Murayama, E.; Otaka, A., New Strategy for the Chemical Synthesis of Proteins. *Tetrahedron* **1988,** *44,* 805–819.

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