JOC The Journal of Organic Chemistry

Article

pubs.acs.org/joc

Artificial Macrocycles by Ugi Reaction and Passerini Ring Closure

Eman M. M. Abdelraheem,[†,‡] Katarzyna Kurpiewska,[§] Justyna Kalinowska-Tłuścik,[§] and Alexander Dömling*[,†]

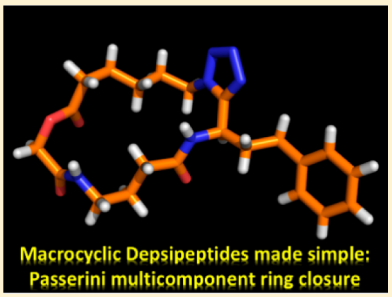
[†]Department of Drug Design, University of Groningen, A. Deusinglaan 1, 9713 AV Groningen, The Netherlands

[‡]Chemistry Department, Faculty of Science, Sohag University, Sohag 82524, Egypt

[§]Faculty of Chemistry, Jagiellonian University, 3 Ingardena Street, 30-060 Kraków, Poland

Supporting Information

ABSTRACT: Artificial macrocycles can be convergently synthesized by a sequence of an Ugi multicomponent reaction (MCR) followed by an intramolecular Passerini MCR used to close the macrocycle. Significantly, in this work, the first intramolecular macrocyclization through a Passerini reaction is described. We describe 21 macrocycles of a size of 15−20. The resulting macrocyclic depsipeptides are model compounds for natural products and could find applications in drug discovery.



INTRODUCTION

Macrocyclic depsipeptides, although smaller in number than their twins, macrocyclic peptides, are an important class of natural products generally associated with interesting biological activity.[1] In depsipeptides, one amide group of a peptide is exchanged by an ester group. Depsipeptides, therefore, are synthetically more demanding due to the α-hydroxycarboxylic acid building blocks and the respective coupling chemistry.[2] A very much underappreciated chemistry toward depsipeptides is the Passerini three-component reaction (3-CR), in which one step gives access to the framework of α-acylhydroxyamides and, surprisingly, few applications are published (Scheme 1).[3−8] Especially with the advent of efficient enantioselective Passerini multicomponent reactions (MCRs), a bright future of this MCR for natural product synthesis can be predicted.[9−12] Again, surprisingly, Passerini MCR has been only described three times in the synthesis of macrocycles and never as a intramolecular macro-ring-closing method.[6,13,14] Recently, artificial macrocycles have become an important target group in organic synthesis due to their usefulness as drug candidates for difficult protein targets, such as protein−protein interactions.[15,16] However, the rules rendering cyclic peptides cell-permeable are far from being understood.[17] The conformational space of depsipeptides is different from that of their peptide analogues due to the second amide → ester exchange, which makes some intra- and intermolecular hydrogen bonding impossible.[18,19]

Thus, it can be speculated that depsipeptides have a higher tendency to penetrate biological membranes, which can be beneficial for target occupancy, oral bioavailability, and organ penetration.[20] On the other hand, ester groups are often easier to cleave enzymatically or spontaneously than amide groups, although some ester groups exhibit decent hydrolysis resistance and are of sufficient metabolic stability to be used as drugs.

RESULTS AND DISCUSSION

The aim of the described work was to provide a fast, efficient, and general method for obtaining artificial macrocyclic depsipeptides using a union of the MCR concept.[21] The novelty of the work includes the first use of the Passerini reaction to ring close macrocycles intramolecularely using bifunctional isocyanocarboxylate (Scheme 1). Topologically, there exist three pathways to close a ring by a Passerini 3-CR (Figure 1), by reacting either an α-isocyano-ω-carboxylic acid and an oxo component (A), an α-oxo-ω-carboxylic acid and an isocyanide (B), and an α-oxo-ω-isocyanide and carboxylic acid component (C).

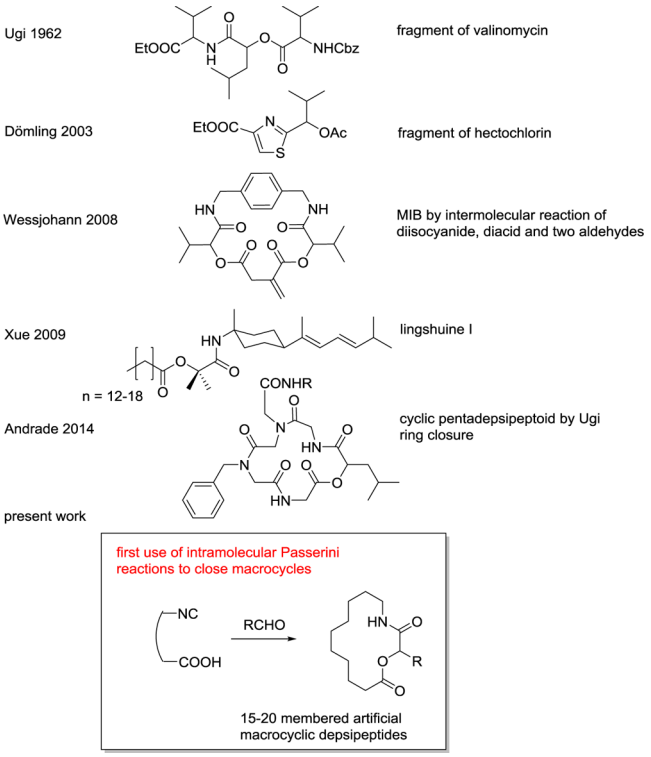
Having convenient access to α-isocyano-ω-carboxylates with different size and substitution pattern, as recently described by us, we first focused here on pathway B.[22] The envisioned synthetic pathway is sketched in Scheme 2.

First, to introduce linkers of multiple chain lengths and side chains, we reacted an oxo component (aldehyde, ketone) **1** with tritylamine **2**, $TMSN_3$ **3**, and a variety of α-isocyano-ω-carboxylic acid methylesters **4** in an Ugi tetrazole MCR to yield **5**. After detritylation, the potassium salt of an α-isocyano-ω-carboxylic acid **7** was coupled to the primary amine **6**, followed by saponification of **9**. Finally, with the help of another oxo component **10**, the macrocycle was closed by a Passerini 3-CR **11**. To increase diversity and to vary ring size, we synthesized eight α-isocyano-ω-carboxylate linkers **9** of different lengths

Received: June 14, 2016
Published: September 6, 2016

DOI: 10.1021/acs.joc.6b01430
J. Org. Chem. 2016, 81, 8789−8795

Scheme 1. Key Contributions to the Use of the Passerini 3-CR in Depsipeptide Synthesis and the Described Work Herein



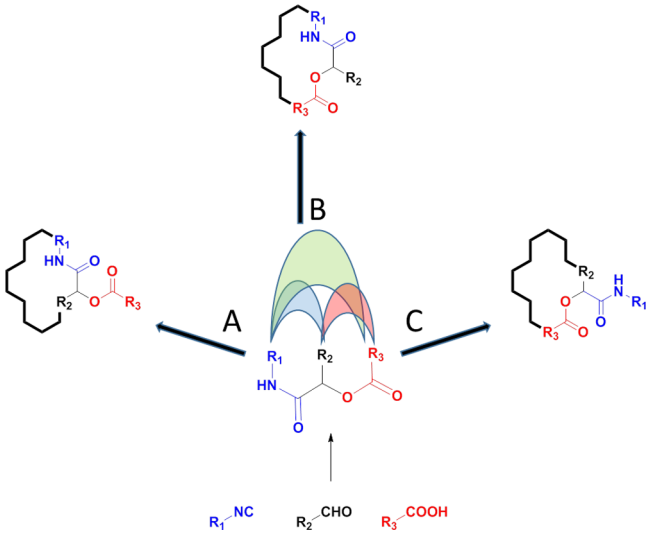
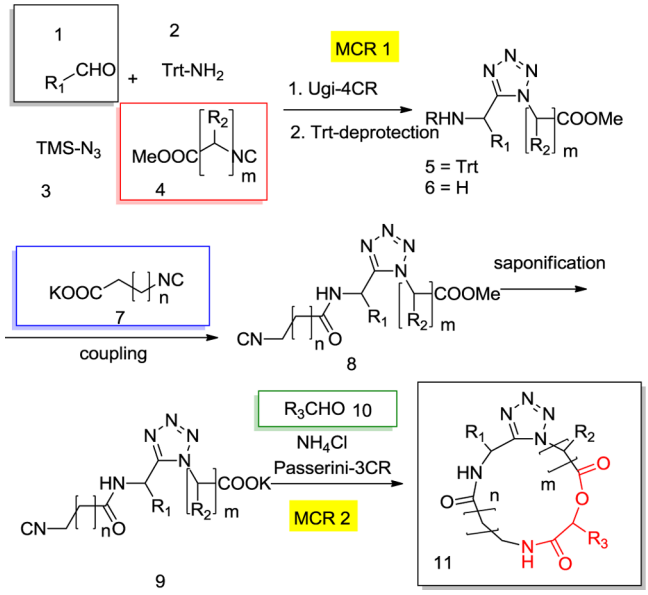


Figure 1. Three topological pathways to form macrocycles using Passerini 3-CR as the ring closure method.

from their commercial starting materials according to Scheme 1 (Supporting Information, S-6).

While all reactions in Scheme 2 have precedence and are well described in the literature, the Passerini macrocyclization is new and required a lot of optimization, including time, solvent, and concentration (Supporting Information Table 1). Surprisingly, after extensive optimization, we found that 0.01 M dilution in water was optimal. The latter aspect is unusual for the Passerini reaction, which is described as performing best in aprotic apolar solvents such as ether, THF, and DCM.[8] The carboxylate might benefit from water in several aspects: it might enhance the

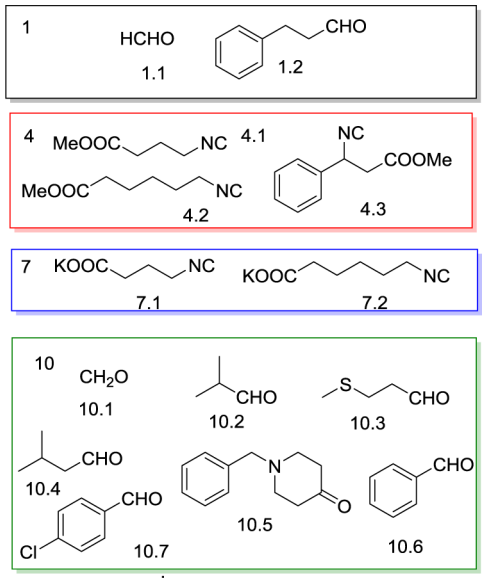
Scheme 2. Synthesis Strategy toward Artificial Macrocyclic Depsipeptides



solubility of the starting material carboxylate as well as the additive used. Most compounds show rotamers in the NMR spectra, and 11.18 is a ~2:3 mixture of two diastereomers, which we could not separate by silica chromatography.

To investigate the scope and limitations, we synthesized a total of 21 examples according to Scheme 3. These examples

Scheme 3. Starting Material Classes Used (Colors Refer to Scheme 2)



exemplify some of the diversity of starting materials that can be used in this four-step sequence to provide macrocyclic depsipeptides (Scheme 4). The ring size was modified from 15 to 20. The yields of all reactions are summarized in Table 1, and interestingly, it was found that they do not depend on the ring sizes. Next, the scope of the Ugi reaction was examined. Two aldehydes **1** and three isocyanides **4** were used to produce five different variations of **5** in good to excellent yields (59–96%, Table 1). Next, the investigation of the Passerini

DOI: 10.1021/acs.joc.6b01430
J. Org. Chem. 2016, 81, 8789–8795

Scheme 4. Structures of the Synthesized Macrocyclic Depsipeptides

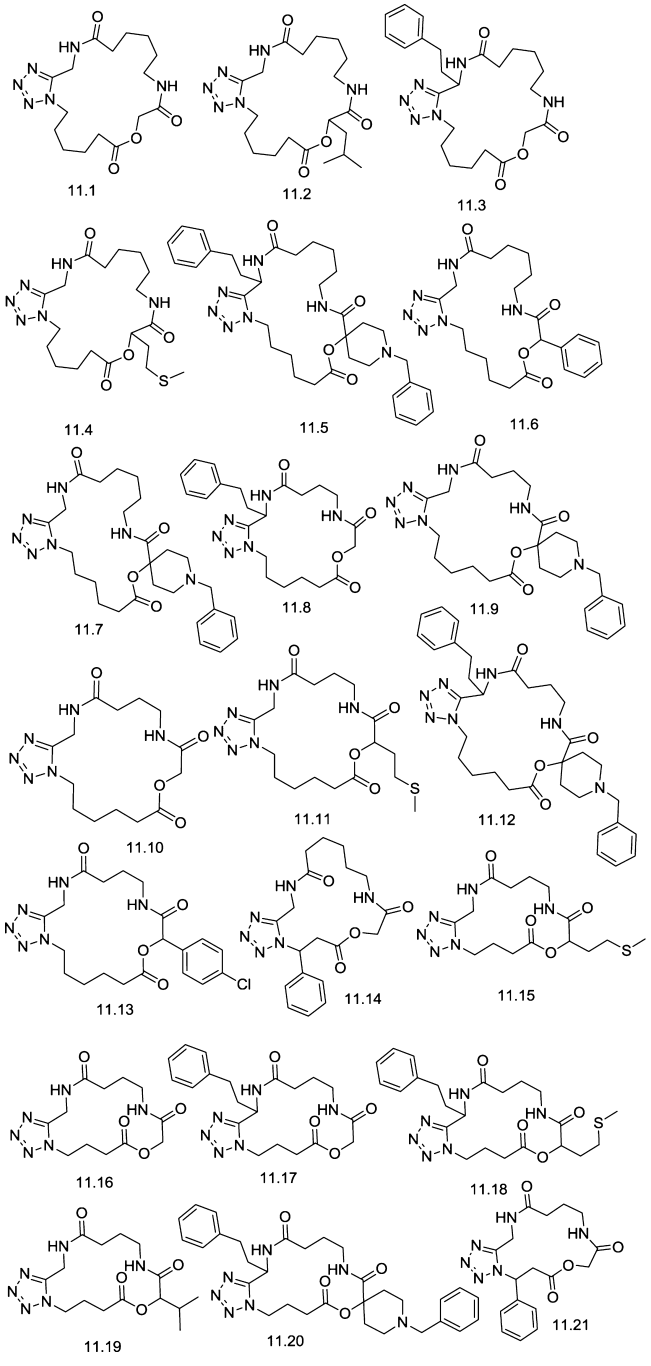


Table 1. Yields of Ugi, Detritylation, Coupling, and Macrocyclization Products

		yield (%)			
entry	educts	**5**	**6**	**8**	**11**
1	1.1, 4.2, 7.2, 10.1	78	88	74	27
2	1.1, 4.2, 7.2, 10.4	78	88	74	20
3	1.2, 4.2, 7.2, 10.1	59	67	56	28
4	1.1, 4.2, 7.2, 10.3	78	88	74	28
5	1.2, 4.2, 7.2, 10.5	59	67	56	30
6	1.1, 4.2, 7.2, 10.6	78	88	74	20
7	1.1, 4.2, 7.2, 10.5	78	88	74	21
8	1.2, 4.2, 7.1, 10.1	59	67	50	25
9	1.1, 4.2, 7.1, 10.5	78	88	75	31
10	1.1, 4.2, 7.1, 10.1	78	88	75	36
11	1.1, 4.2, 7.1, 10.3	78	88	75	32
12	1.2, 4.2, 7.1, 10.5	59	67	50	20
13	1.1, 4.2, 7.1, 10.7	78	88	75	21
14	1.1, 4.3, 7.2, 10.1	90	80	64	28
15	1.1, 4.1, 7.1, 10.3	96	80	64	35
16	1.1, 4.1, 7.1, 10.1	96	80	64	25
17	1.2, 4.1, 7.1, 10.1	59	75	56	29
18	1.2, 4.1, 7.1, 10.3	59	75	56	25
19	1.1, 4.1, 7.1, 10.2	96	80	64	35
20	1.2, 4.1, 7.1, 10.5	59	75	56	21
21	1.1, 4.3, 7.1, 10.1	90	80	73	20

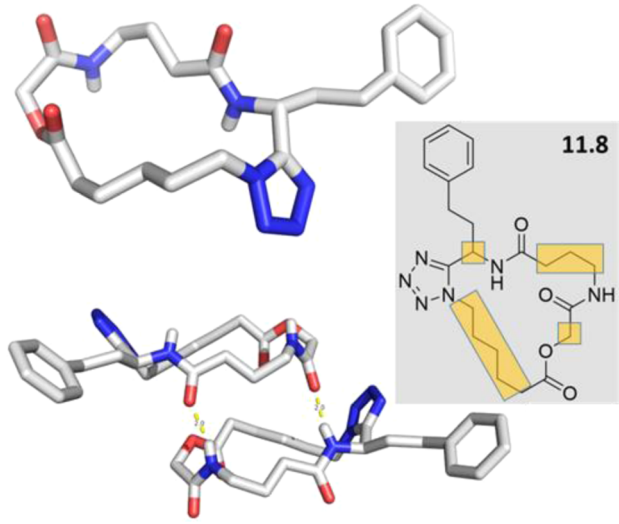


Figure 2. Representative MCR-derived depsipeptidic 18-membered macrocycle **11.8** in the solid state. Top: View on top of the ring plane. Bottom: Two adjacent antiparallel stacked macrocycles forming two short hydrogen bonds (2 Å, yellow dotted lines) involving secondary amide groups. The flexible elements of the macrocycle in the box are shown in orange. Rendering using PyMol.

cyclization step using several commercially available aliphatic, aromatic, and heterocyclic oxo components **10** as aldehydes and ketones yielded macrocyclic depsipeptides **11** in low to moderate yield after purification by column chromatography (20−36%, Table 1). We found that the presence of the NH_4Cl additive is necessary for the Passerini ring closure, likely due to the use of the potassium salt in the linkers **9**, for neutralization purposes.

We were also able to grow crystals of macrocycle **11.8** that were useful for X-ray structure analysis (Figure 2). This structure gives some insight into the intra- and intermolecular interactions in the solid state. For example, it can be observed that the two secondary amides form intermolecular hydrogen bondings to a neighbor macrocycle, whereas the *cis*-amide bioisosteric tetrazole moiety is not involved with hydrogen bonding. Looking into the different modules of **11.8**, one can define the two amide groups, the tetrazole and the lactone group, as rigid elements which are separated by flexible sp^3 center-based C1, C3, and C5 chain elements. These linker fragments ultimately will determine the flexibility of the overall macrocyclic conformations in aqueous and lipophilic environments, which will be a determinant of the passive diffusion through cell membranes.[18]

DOI: 10.1021/acs.joc.6b01430
J. Org. Chem. 2016, 81, 8789−8795

CONCLUSIONS

In conclusion, we describe here the first use of the Passerini reaction to close macrocycles and thus form artificial macrocyclic depsipeptides. This is meaningful because macrocyclic depsipeptides are a large group of highly bioactive natural products. The overall sequence used here to introduce different ring sizes and side chain variations is just four steps using readily available starting materials. Thus, we foresee multiple applications for these artificial macrocycles as unusual scaffolds to target difficult protein−protein interactions and other postgenomic targets. Libraries of such macrocyclic depsipeptides are currently screened in our laboratory for biological activity and will be reported in due course. This novel method will add to the tool kit of macrocyclizations by MCR.[23−26]

EXPERIMENTAL SECTION

General Methods. All chemicals were purchased from commercial suppliers and used without any purification unless otherwise noted. Nuclear magnetic resonance spectra were recorded. Chemical shifts for ^{1}H NMR were reported as δ values, and coupling constants were in hertz (Hz). The following abbreviations were used for spin multiplicity: s = singlet, bs = broad singlet, d = doublet, t = triplet, q = quartet, quin = quintet, dd = double of doublets, ddd = double of doublet of doublets, m = multiplet. Chemical shifts for ^{13}C NMR reported in parts per million (ppm) relative to the solvent peak. Thin layer chromatography was performed on silica gel plates (0.20 mm thick, particle size 25 μm). Flash chromatography was performed using RediSep R_f normal-phase silica flash columns (silica gel 60 Å, 230−400 mesh). All microwave irradiation reactions were carried out in a Biotage Initiator microwave synthesizer with an external infrared sensor type. Electrospray ionization mass spectra (ESI-MS) and high-resolution mass spectra were recorded.

Procedure and Analytical Data for *N*-Trityl-Protected α-Aminotetrazoles. The synthesis of the *N*-trityl-protected α-aminotetrazole was based on a previously published procedure for *N*-trityl-protected α-aminotetrazole.[22] Briefly, aldehyde (1.5 mmol) and tritylamine (1.0 mmol) were suspended in MeOH (1 mL) in a sealed vial with a magnetic stirring bar. The reaction was heated at 100 °C for 15 min using microwave irradiation. Then isocyanide (1.0 mmol) and azidotrimethylsilane (1.0 mmol) were added into the reaction mixture and further irradiated at 100 °C for 15 min. The solvent was removed under reduced pressure, and the residue was purified using flash chromatography (petroleum ether/ethyl acetate 1:1).

Methyl 6-(5-((Tritylamino)methyl)-1H-tetrazol-1-yl)hexanoate ***5.1****, Methyl 6-(5-(3-Phenyl-1-(tritylamino)propyl)-1H-tetrazol-1-yl)-hexanoate* ***5.3****, Methyl 4-(5-((Tritylamino)methyl)-1H-tetrazol-1-yl)butanoate* ***5.15****, and Methyl 4-(5-(3-Phenyl-1-(tritylamino)-propyl)-1H-tetrazol-1-yl)butanoate* ***5.17****.* ^{1}H NMR and ^{13}C NMR signals are in agreement with the literature data.[22]

Methyl 3-Phenyl-3-(5-((tritylamino)methyl)-1H-tetrazol-1-yl)-propanoate ***5.14****.* The product was obtained as a white solid (90%, 0.453 g): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.47 (d, J = 7.8, 6H), 7.34−7.28 (m, 9H), 7.25 (t, J = 7.2, 3H), 7.12−7.08 (m, 2H), 5.92 (dd, J = 10.3, 4.6, 1H), 3.82 (dd, J = 17.4, 10.3, 1H), 3.72 (dd, J = 14.0, 7.9, 1H), 3.65 (s, 3H), 3.65−3.61 (m, 1H), 3.14 (dd, J = 17.4, 4.6, 1H), 2.47 (t, J = 7.7, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 170.0, 153.9, 144.6, 136.8, 129.3, 129.1, 128.6, 128.1, 126.8, 126.6, 71.1, 58.5, 52.3, 40.2, 36.9 ppm; MS (ESI) m/z calcd $[M + H]^+$ 504.59; found $[M + Na]^+$ 527.19.

Procedure and Analytical Data for N-Deprotected α-Aminotetrazoles. The synthesis of the N-deprotected α-aminotetrazole was based on a previously published procedure for N-deprotected α-aminotetrazole.[22] *N*-Trityl-protected α-aminotetrazole (0.5 mmol) was dissolved in DCM (2.5 mL) in a vial with a magnetic stirring bar. Trifluoroacetic acid (1.0 mmol, 77 μL) was added dropwise. The reaction was developed for 1 min and was purified using a silica pad wetted with heptane/EtOAc (1:1). The side product was washed out with heptane/EtOAc (1:1). The N-deprotected α-aminotetrazole was collected with CH_2Cl_2/MeOH (1:1). The solvent was removed under reduced pressure, which gave the pure product.

Methyl 6-(5-(Aminomethyl)-1H-tetrazol-1-yl)hexanoate ***6.1****, Methyl 6-(5-(1-Amino-3-phenylpropyl)-1H-tetrazol-1-yl)hexanoate* ***6.3****, Methyl 4-(5-(Aminomethyl)-1H-tetrazol-1-yl)butanoate* ***6.15****, and Methyl 4-(5-(1-Amino-3-phenylpropyl)-1H-tetrazol-1-yl)-butanoate* ***6.17****.* ^{1}H NMR and ^{13}C NMR signals are in agreement with the literature data.[22]

Methyl 3-(5-(Aminomethyl)-1H-tetrazol-1-yl)-3-phenylpropanoate ***6.14****.* The product was obtained as a white solid (80%, 0.299 g): ^{1}H NMR (500 MHz, DMSO) δ 8.86 (s, 2H), 7.51 (br, 2H), 7.46−7.35 (m, 3H), 6.34−6.15 (m, 1H), 4.61 (dd, J = 16.5, 5.0, 1H), 4.45 (dd, J = 16.4, 4.8, 1H), 3.67−3.61 (m, 1H), 3.57 (s, 3H), 3.51−3.45 (m, 1H); ^{13}C NMR (126 MHz, DMSO) δ 170.5, 151.0, 136.8, 129.5, 129.4, 127.9, 57.9, 52.4, 39.2, 33.2 ppm; MS (ESI) m/z calcd $[M + H]^+$ 262.28; found $[M + H]^+$ 262.01.

Procedure and Analytical Data for the Coupling Reactions. A suspension of N-deprotected α-aminotetrazole derivatives (1.0 mmol), potassium isocyanide derivatives (1.2 mmol), and triethylamine (1.0 mmol) in CH_3CN (10 mL) were stirred for 10 min at 0 °C. Next, HOBt (1.0 mmol) and DCC (1.0 mmol) were added to the mixture, and the reaction mixture was stirred for 48 h. The insoluble materials were filtered off, and the filtrate was evaporated. The residue was purified by column chromatography (petroleum ether/ethyl acetate 1:4).

Methyl 6-(5-((6-Isocyanohexanamido)methyl)-1H-tetrazol-1-yl)-hexanoate ***8.1****, Methyl 6-(5-(1-(6-Isocyanohexanamido)-3-phenylpropyl)-1H-tetrazol-1-yl)hexanoate* ***8.3****, Methyl 6-(5-(1-(4-Isocyanobutanamido)-3-phenylpropyl)-1H-tetrazol-1-yl)hexanoate* ***8.8****, Methyl 6-(5-((4-Isocyanobutanamido)methyl)-1H-tetrazol-1-yl)-hexanoate* ***8.9****, and Methyl 4-(5-((4-Isocyanobutanamido)-methyl)-1H-tetrazol-1-yl)butanoate* ***8.15****.* ^{1}H NMR and ^{13}C NMR signals are in agreement with the literature data.[22]

Methyl 3-(5-((6-Isocyanohexanamido)methyl)-1H-tetrazol-1-yl)-3-phenylpropanoate ***8.14****.* The product was obtained as a yellow oil (64%, 0.246 g): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.43−7.39 (m, 2H), 7.38−7.33 (m, 3H), 7.29 (t, J = 5.7, 1H), 6.25 (dd, J = 10.8, 4.1, 1H), 4.78 (dd, J = 5.9, 2.1, 2H), 3.79 (dd, J = 17.6, 10.8, 1H), 3.61 (s, 3H), 3.42−3.32 (m, 2H), 3.15 (dd, J = 17.6, 4.1, 1H), 2.26−2.21 (m, 2H), 1.71−1.59 (m, 4H), 1.50−1.39 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 173.2, 169.9, 155.8, 152.8, 136.6, 129.3, 129.2, 126.9, 58.3, 52.2, 41.4, 40.1, 35.6, 31.7, 28.8, 25.8, 24.3 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{19}H_{25}N_6O_3$ 385.1986; found 385.1983.

Methyl 3-(5-((4-Isocyanobutanamido)methyl)-1H-tetrazol-1-yl)-3-phenylpropanoate ***8.21****.* The product was obtained as a yellow oil (73%, 0.259 g): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.44 (t, J = 5.7, 1H), 7.41−7.32 (m, 5H), 6.23 (dd, J = 10.8, 4.0, 1H), 4.78−4.73 (m, 2H), 3.79 (dd, J = 17.7, 10.8, 1H), 3.61 (s, 3H), 3.46 (dd, J = 8.9, 4.1, 2H), 3.15 (dd, J = 17.6, 4.0, 1H), 2.39−2.34 (m, 2H), 1.99−1.97 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 171.9, 170.0, 161.9, 152.7, 136.5, 129.3, 129.2, 126.9, 58.4, 52.3, 41.0, 40.0, 31.8, 31.7, 24.5 ppm; HRMS (ESI/TOF-Q) m/z $[M + H + H_2O]^+$ calcd for $C_{17}H_{23}N_6O_4$ 375.1781; found 375.1779.

General Procedure for the Saponification Reactions. The isocyanide ester (1.0 mmol) was dissolved in EtOH (1 mL), and potassium hydroxide (1.5 mmol) was added. The reaction was stirred at room temperature. After consumption of the starting material as indicated by TLC, the solvent was removed under vacuum and the potassium salt was subjected directly to the next step.

Procedure and Analytical Data for the Macrocyclization Reaction. A mixture of α-isocyano-ω-carboxylic acid (1.0 mmol) and ammonium chloride (1.5 mmol) in H_2O (0.01 M, 100 mL) was stirred at room temperature for 30 min, and then aldehyde or ketone (1.0 mmol) was added to the reaction mixture and further stirred for 72 h. The solvent was removed under reduced pressure, and the residue was purified using flash chromatography (CH_2Cl_2/MeOH 9:1).

6,7,8,9,14,15,16,17,18,19,21,22-Dodecahydro-5H-tetrazolo[5,1-m][1,4,11,14]oxatriazacycloicosine-10,13,20(12H)-trione ***(11.1)****.* The product was obtained as a white solid (27%, 0.098g, mp 194−196 °C): ^{1}H NMR (500 MHz, DMSO) δ 8.52 (t, J = 5.6, 1H), 7.76 (t, J = 5.7, 1H), 4.60 (d, J = 4.1, 2H), 4.39 (s, 2H), 4.29 (t, J = 7.5, 2H),

DOI: 10.1021/acs.joc.6b01430
J. Org. Chem. 2016, 81, 8789−8795

3.13−3.09 (m, 2H), 2.41 (t, J = 6.8, 2H), 2.11 (t, J = 7.0, 2H), 1.87−1.77 (m, 2H), 1.64−1.55 (m, 2H), 1.48−1.44 (m, 2H), 1.43−1.37 (m, 2H),1.35−1.29 (m, 2H), 1.20−1.16 (m, 2H); ^{13}C NMR (126 MHz, DMSO) δ 173.0, 172.9, 167.3, 153.3, 63.1, 46.9, 37.8, 35.1, 33.2, 31.9, 29.1, 28.7, 25.6, 25.2, 24.3, 24.1 ppm; HRMS (ESI/TOF-Q) m/z [M + H]$^+$ calcd for $C_{16}H_{27}N_6O_4$ 367.20874; found 367.20883.

*12-Isobutyl-6,7,8,9,14,15,16,17,18,19,21,22-dodecahydro-5H-tetrazolo[5,1-m][1,4,11,14]oxatriazacycloicosine-10,13,20-(12H)-trione (**11.2**).* The product was obtained as a white solid (20%, 0.084 g, mp 200−202 °C): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.41 (s,1H), 4.76−4.56 (m, 2H), 4.42−4.39 (m, 2H), 4.10−4.06 (m, 1H), 3.23−3.13 (m, 2H), 2.36−2.28 (m, 2H), 2.23−2.21 (m, 2H), 1.98−1.88 (m, 2H), 1.65−1.56 (m, 5H), 1.52−1.42 (m, 2H), 1.37−1.35 (m, 2H), 1.33−1.25 (m, 2H), 1.23−1.21 (m, 1H), 0.94 (d, J = 6.6, 2H), 0.90 (dd, J = 6.5, 3.3, 3H), 0.86 (d, J = 6.1,1H); ^{13}C NMR (126 MHz, DMSO) δ 172.9, 172.4, 153.2, 51.9, 51.6, 47.0, 40.9, 38.1, 35.1, 32.1, 29.4, 28.9, 25.7, 25.4, 24.5, 23.3, 22.3 ppm; HRMS (ESI/TOF-Q) m/z [M + H]$^+$ calcd for $C_{20}H_{35}N_6O_4$ 423.27143; found 423.27143.

*22-Phenethyl-6,7,8,9,14,15,16,17,18,19,21,22-dodecahydro-5H-tetrazolo[5,1-m][1,4,11,14]oxatriazacycloicosine-10,13,20-(12H)-trione (**11.3**).* The product was obtained as a white solid (28%, 0.132 g, mp 157−159 °C): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.28 (t, J = 7.3, 2H), 7.21 (t, J = 7.4, 1H), 7.11 (d, J = 7.5, 2H), 6.54 (s, 1H), 6.27 (d, J = 8.5, 1H), 5.28−5.24 (m, 1H), 4.61 (d, J = 15.5, 1H), 4.51 (d, J = 15.5, 1H), 4.34−4.28 (m, 1H), 4.00−88 (m, 1H), 3.50−3.38 (m, 1H), 3.36−3.22 (m, 1H), 2.66 (t, J = 7.0, 2H), 2.39 (t, J = 7.5, 2H), 2.36−2.29 (m, 1H), 2.28−2.23 (m, 1H), 2.19 (m, 2H), 1.92−1.74 (m, 4H), 1.68−1.62 (m, 2H), 1.61−1.56 (m, 2H), 1.50−1.40 (m, 2H), 1.37−1.30 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ= 173.1, 172.1, 167.5, 155.9, 140.0, 129.3, 128.8, 127.2, 63.5, 47.1, 42.1, 38.6, 36.0, 35.3, 33.6, 32.1, 28.9, 28.2, 26.1, 25.4, 24.0, 23.7 ppm; HRMS (ESI/TOF-Q) m/z [M + H]$^+$ calcd for $C_{24}H_{34}N_6O_4$ 471.27143; found 471.27139.

*12-(2-(Methylthio)ethyl)-6,7,8,9,14,15,16,17,18,19,21,22-dodecahydro-5H-tetrazolo[5,1-m][1,4,11,14]oxatriazacycloicosine-10,13,20(12H)-trione (**11.4**).* The product was obtained as a white solid (28%, 0.123 g, mp 158−200 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.46−7.40 (bs, 1H), 4.70 (t, J = 6.2, 1H), 4.65 (d, J = 5.9, 2H), 4.43−4.37 (m, 3H), 3.36−3.18 (m, 2H), 2.55−2.46 (m, 3H), 2.30−2.22 (m, 4H), 2.11−2.09 (m, 2H), 2.06−2.05 (m, 2H), 2.01 (s, 3H), 1.99−1.95 (m, 1H), 1.90−1.86 (m, 3H), 1.75−1.66 (m, 1H), 1.64−1.56 (m,45H), 1.53−1.45 (m, 2H), 1.38−1.31 (m, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.0, 173.7, 172.6, 152.9, 73.3, 47.8, 38.7, 35.3, 34.2, 32.1, 31.7, 29.7, 28.3, 26.2, 25.4, 24.5, 23.8, 15.7 ppm; HRMS (ESI/TOF-Q) m/z [M − H]$^+$ calcd for $C_{19}H_{31}N_6O_4S$ 439.2133; found 439.21298.

*1-Benzyl-22′-phenethyl-6′,7′,8′,9′,14′,15′,16′,17′,18′,19′,21′,22′-dodecahydrospiro[piperidine-4,12′-tetrazolo[5,1-m][1,4,11,14]-oxatriazacycloicosine]10′,13′,20′(5′H)-trione (**11.5**).* The product was obtained as a white solid (30%, 0.189 g, mp 168−170 °C): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.34−7.27 (m, 4H), 7.27−7.22 (m, 3H), 7.18−7.16 (m, 1H), 7.13−7.09 (m, 2H), 6.81 (d, J = 9.0, 1H), 6.15 (t, J = 5.5, 1H), 5.38−5.29 (m, 1H), 4.34−4.17 (m, 2H), 4.14−4.01 (m, 1H), 3.52 (s, 2H), 3.27−3.24 (m, 2H), 2.81−2.69 (m, 2H), 2.65 (t, J = 7.4, 2H), 2.37−2.20 (m, 6H), 2.20−2.04 (m, 4H), 1.94−1.89 (m, 1H), 1.84−1.77 (m, 1H), 1.67−1.48 (m, 4H), 1.44−1.41 (m, 2H), 1.39−1.21 (m, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 173.5, 172.2, 171.9, 155.5, 140.2, 137.5, 129.5, 128.9, 128.6, 128.5, 127.6, 126.8, 79.9, 62.7, 48.8, 48.7, 47.0, 42.0, 38.6, 35.3, 35.1, 34.0, 32.1, 31.3, 28.6, 28.2, 25.8, 25.2, 23.7, 23.6 ppm; HRMS (ESI/TOF-Q) m/z [M + H]$^+$ calcd for $C_{35}H_{48}N_7O_4$ 630.37623; found 630.37634.

*12-Phenyl-6,7,8,9,14,15,16,17,18,19,21,22-dodecahydro-5H-tetrazolo[5,1-m][1,4,11-,14]oxatriazacycloicosine-10,13,20-(12H)-trione (**11.6**).* The product was obtained as a yellow oil (20%, 0.088 g): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.45 (d, J = 7.3, 2H), 7.42−7.31 (m, 3H), 6.52 (s, 1H), 4.80−4.58 (m, 2H), 4.50−4.41 (m, 2H), 3.40−3.32 (m, 1H), 3.25−2.23 (m, 1H), 2.32 (dd, J = 8.3, 6.3, 2H), 2.24−2.21 (m, 2H), 1.98−1.93 (m, 2H), 1.70−1.61 (m, 4H), 1.54−1.46 (m, 2H), 1.45−1.42 (m, 2H), 1.31−1.28 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 173.7, 173.6, 172.9, 152.3, 129.2, 128.9, 128.8, 126.9, 74.4, 47.6, 39.1, 35.9, 34.1, 32.2, 29.6, 29.2, 26.1, 25.9, 24.8, 24.4 ppm; HRMS (ESI/TOF-Q) m/z [M − H]$^+$ calcd for $C_{22}H_{29}N_6O_4$ 441.22558; found 441.22568.

*1-Benzyl-6′,7′,8′,9′,14′,15′,16′,17′,18′,19′,21′,22′-dodecahydrospiro[piperidine-4,12′-tetrazolo[5,1-m][1,4,11,14]oxatriazacycloicosine]-10′,13′,20′(5′H)-trione (**11.7**).* The product was obtained as brown oil (21%, 0.110 g). A mixture of rotamers was observed, and the majority of rotamers was taken: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.36 (d, J = 6.8, 2H), 7.33−7.29 (m, 4H), 7.27 (d, J = 6.2, 3H), 4.66 (dd, J = 5.7, 3.6, 1H), 4.41 (dd, J = 7.3, 3.2, 1H), 3.70−3.66 (m, 2H), 3.60 (d, J = 12.5 1H), 3.45 (d, J = 12.7, 1H), 3.30−3.17 (m, 1H), 3.13 (d, J = 11.4, 1H), 3.03−2.94 (m, 1H), 2.78−2. 71 (m, 3H), 2.66−2.49 (m, 4H), 2.49−2.42 (m, 1H), 2.33−2.10 (m, 4H), 1.91 (dd, J = 14.9, 7.5, 1H), 1.77 (bs,1H), 1.62−1.58 (m, 4H), 1.55−1.38 (m, 3H), 1.40−1.25 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 173.9, 171.9, 171.6, 152.5, 129.9, 128.5, 128.1, 78.7, 61.8, 47.4, 38.4, 35.2, 34.4, 33.7, 31.9, 31.1, 29.2, 28.6, 28.0, 25.7, 24.9, 24.3, 23.5, 22.7 ppm; HRMS (ESI/TOF-Q) m/z [M + H]$^+$ calcd for $C_{27}H_{40}N_7O_4$ 526.31362; found 526.31354.

*20-Phenethyl-6,7,8,9,14,15,16,17,19,20-decahydro-5H-tetrazolo-[5,1-k][1,4,9,12] oxatriazacyclooctadecene-10,13,18(12H)-trione (**11.8**).* The product was obtained as a white solid (25%, 0.110 g, mp 171−173 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.29 (s, 1H), 7.34−7.28 (m, 3H), 7.13 (d, J = 7.5, 2H), 5.27 (m, 1H), 4.60 (s, 2H), 4.47−4.34 (m, 1H), 4.07−3.91 (m, 1H), 3.44−3.30 (m, 2H), 2.73 (t, J = 7.1, 2H), 2.57−2.47 (m, 2H), 2.45−2.39 (m, 2H), 2.36−2.33 (m, 2H), 2.26−2.22 (m, 1H), 1.97−1.88 (m, 1H), 1.89−1.82 (m, 2H), 1.76−1.67 (m, 2H), 1.48−1.39 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.3, 171.8, 167.5, 155.4, 139.3, 128.8, 128.3, 126.7, 62.6, 46.6, 41.6, 40.2, 34.7, 34.7, 32.5, 31.5, 27.7, 24.3, 22.7, 22.2 ppm; HRMS (ESI/TOF-Q) m/z [M + H]$^+$ calcd for $C_{22}H_{31}N_6O_4$ 443.24013; found 443.23993.

*1-Benzyl-6′,7′,8′,9′,14′,15′,16′,17′,19′,20′-decahydrospiro-[piperidine-4,12′-tetrazolo-[5,1-k][1,4,9,12]oxatriazacyclooctadecene]-10′,13′,18′(5′H)-trione (**11.9**).* The product was obtained as a white solid (31%, 0.154g, mp 162−164 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.95 (t, J = 5.7, 1H), 7.70 (t, J = 5.7, 1H), 7.32 (d, J = 7.3, 4H), 7.29−7.28 (m, 1H), 7.20 (t, J = 5.3, 1H), 4.79 (d, J = 5.9, 2H), 4.69 (d, J = 5.9, 2H), 4.42 (t, J = 7.0, 3H), 4.30 (t, J = 6.8, 2H), 3.77 (s, 2H), 3.46 (t, J = 6.3, 2H), 3.33−3.27 (m, 2H), 3.06−2.97 (m, 2H), 2.52 (t, J = 10.3, 2H), 2.45 (t, J = 7.1, 2H), 2.37−2.31 (m, 4H), 2.29 (t, J = 7.0, 3H), 2.20−2.16 (m, 2H), 1.98−1.95 (m, 4H), 1.92−1.90 (m, 3H), 1.78−1.75 (m, 2H), 1.66−1.59 (m, 3H), 1.57−1.53 (m, 2H), 1.37−1.30 (m, 3H), 1.30−1.25 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.1, 172.4, 172.1, 152.4, 130.2, 129.9, 128.6, 128.2, 78.7, 61.8, 48.1, 47.4, 41.0, 38.9, 34.2, 33.2, 32.0, 31.8, 31.1, 29.1, 28.5, 25.6, 25.2, 24.9, 24.5, 24.2, 23.3 ppm; HRMS (ESI/TOF-Q) m/z [M − H]$^+$ calcd for $C_{25}H_{34}N_7O_4$ 496.26778; found 496.26785.

*6,7,8,9,14,15,16,17,19,20-Decahydro-5H-tetrazolo[5,1-k][1,4,-9,12]oxatriazacyclooctadecene-10,13,18(12H)-trione (**11.10**).* The product was obtained as a white solid (36%, 0.121g, mp 153−155 °C): ^{1}H NMR (500 MHz, DMSO) δ 8.65 (t, J = 5.6, 1H), 8.03 (t, J = 5.1,1H), 4.61 (d, J = 5.7, 2H), 4.41 (s, 2H), 4.30 (t, J = 7.2, 2H), 3.15 (d, J = 5.5, 2H), 2.43 (t, J = 6.5, 2H), 2.15 (t, J = 7.0, 2H), 1.90−1.80 (m, 2H), 1.66−1.55 (m, 4H),1.32−1.23 (m, 2H); ^{13}C NMR (126 MHz, DMSO) δ 173.1, 172.5, 167.2, 153.1, 62.9, 46.8, 37.8, 32.7, 32.2, 31.6, 28.6, 24.7, 24.1, 23.5 ppm; HRMS (ESI/TOF-Q) m/z [M + H]$^+$ calcd for $C_{14}H_{23}N_6O_4$ 339.17753; found 339.17746.

*12-(2-(Methylthio)ethyl)-6,7,8,9,14,15,16,17,19,20-decahydro-5H-tetrazolo[5,1-k][1,4,9,12]oxatriazacyclooctadecene-10,13,18-(12H)-trione (**11.11**).* The product was obtained as a white solid (32%, 0.132 g, mp 196−198 °C): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.61 (s, 1H), 7.53 (s, 1H), 5.22 (m, 1H), 4.74 (d, J = 6.0, 2H), 4.50−4.42 (m, 1H), 4.40−4.32 (m, 1H), 3.39−3.34 (m, 1H), 3.33−3.25 (m, 1H), 2.61−2.56 (m, 1H), 2.53−2.44 (m, 3H), 2.41 (t, J = 6.0, 2H), 2.17−2.08 (m, 2H), 2.07 (s, 3H), 2.05−2.03 (m, 1H), 1.97−1.94 (m, 1H), 1.82−1.78 (m, 3H), 1.74−1.69 (m, 1H), 1.49−1.41 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.2, 172.5, 169.9, 152.3, 72.8, 47.1, 42.3, 39.5, 34.0, 32.9, 31.3, 29.6, 28.2, 24.7, 23.8, 22.8, 15.4 ppm; HRMS

DOI: 10.1021/acs.joc.6b01430
J. Org. Chem. 2016, 81, 8789−8795

(ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{17}H_{29}N_6O_4S$ 413.19655; found 413.1965.

*1-Benzyl-20′-phenethyl-6′,7′,8′,9′,14′,15′,16′,17′,19′,20′-decahydrospiro[piperidine-4,12′-tetrazolo[5,1-k][1,4,9,12]-oxatriazacyclooctadecene]-10′,13′,18′(5′H)-trione (**11.12**).* The product was obtained as a yellow solid (20%, 0.120 g, mp 140−142 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.93 (d, J = 9.0, 1H), 7.57 (s, 1H), 7.40−7.33 (m, 5H), 7.28−7.23 (m, 2H), 7.22−7.19 (m,1H), 7.18−7.10 (m, 2H), 5.39−5.31 (m, 1H), 4.29−4.16 (m, 2H), 3.42−2.98 (m, 4H), 2.79 (t, J = 6.1, 2H), 2.77−2.63 (m, 4H), 2.52 (t, J = 6.0, 2H), 2.47−2.23 (m, 8H), 1.98−1.73 (m, 4H), 1.62−1.59 (m, 1H), 1.49−1.44 (m, 1H),1.38−1.17 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.0, 172.2, 171.9, 155.3, 140.3, 140.2, 129.2, 128.9, 128.8, 128.6, 127.6, 126.6, 78.6, 62.1, 48.2, 47.2, 42.4, 41.3, 39.6, 34.9, 33.3, 32.1, 30.6, 28.3, 26.0, 25.0, 23.1 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{33}H_{44}N_7O_4$ 602.34493; found 602.34497.

*12-(4-Chlorophenyl)-6,7,8,9,14,15,16,17,19,20-decahydro-5H-tetrazolo[5,1-k][1,4,-9,12]oxatriazacyclooctadecene-10,13,18(12H)-trione (**11.13**).* The product was obtained as a white solid (21%, 0.094g, mp 206−208 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, DMSO) δ 8.61 (bs, 1H), 7.98 (s, 1H), 7.68−7.66 (m,1H), 7.49−7.37 (m, 4H), 4.57 (d, J = 5.7, 2H), 4.36 (t, J = 7.2, 2H), 3.11−2.98 (m, 2H), 2.19 (t, J = 7.3, 2H), 2.15−2.11 (m, 2H),1.82−1.77 (m, 2H),1.64−1.59 (m, 2H), 1.53−1.48 (m, 2H), 1.30−1.22 (m, 2H); ^{13}C NMR (126 MHz, DMSO) δ 174.8, 172.9, 172.7, 153.5, 137.2, 134.7, 129.7, 129.4, 128.8, 128.6, 56.6, 47.0, 37.2, 33.9, 32.8, 31.9, 29.2, 25.8, 25.6, 24.4 ppm; HRMS (ESI/TOF-Q) m/z $[M - H]^+$ calcd for $C_{20}H_{25}ClN_6O_4$ 447.1553; found 447.1556.

*5-Phenyl-5,6,11,12,13,14,15,16,18,19-decahydrotetrazolo[5,1-m][1,4,11,14]oxatriazacycloheptadecene-7,10,17(9H)trione (**11.14**).* The product was obtained as a white solid (28%, 0.112g, mp 151−153 °C): ^{1}H NMR (500 MHz, MeOD) δ 7.77 (d, J = 16.1, 1H), 7.63−7.59 (m, 2H), 7.43−7.38 (m, 3H), 6.62 (d, J = 16.1, 1H), 4.64 (d, J = 4.9, 4H), 3.34−3.32 (m, 2H), 3.23 (t, J = 7.0, 2H), 2.27 (t, J = 7.5, 2H), 1.67−1.61 (m, 2H), 1.55−1.51 (m, 2H), 1.38−1.33 (m, 2H); ^{13}C NMR (126 MHz, MeOD) δ 177.0, 170.6, 168.1, 156.9, 147.6, 136.2, 130.6, 129.9, 64.1, 40.5, 37.0, 34.5, 30.5, 27.9, 26.7 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{19}H_{25}N_6O_4$ 401.19318; found 401.19305.

*10-(2-(Methylthio)ethyl)-6,7,12,13,14,15,17,18-octahydro-5H-tetrazolo[5,1-k][1,4,9,-12]oxatriazacyclohexadecene-8,11,16(10H)-trione (**11.15**).* The product was obtained as a white solid (35%, 0.134 g, mp 190−192 °C): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.93 (s, 1H), 7.26 (s, 1H), 5.38 (dd, J = 7.5, 4.0, 1H), 4.74−4.64 (m, 1H), 4.52 (dd, J = 15.5, 4.5, 1H), 4.43−4.41 (m, 1H), 4.40−4.37 (m, 1H), 3.38−3.29 (m, 1H), 3.25−3.32 (m, 1H), 2.60−2.53 (m, 2H), 2.52−2.45 (m, 4H), 2.44−2.38 (m, 2H), 2.25−2.19 (m,1H), 2.14−2.08 (m, 1H), 2.07 (s, 3H), 1.86−1.83 (m, 2H), 1.75−1.70 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.9, 172.1, 169.1, 152.9, 72.5, 46.2, 40.2, 34.3, 32.8, 31.3, 29.6, 29.5, 23.5, 22.9, 15.4 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{15}H_{25}N_6O_4S$ 385.16525; found 385.16525.

*6,7,12,13,14,15,17,18-Octahydro-5H-tetrazolo[5,1-k][1,4,9,-12]-oxatriazacyclohexadecene-8,11,16(10H)-trione (**11.16**).* The product was obtained as a white solid (25%, 0.077 g, mp 144−146 °C): ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.16 (s, 1H), 7.89 (t, J = 4.9, 1H), 4.54 (d, J = 9, 2H), 4.49 (d, J = 5.5, 2H), 3.29−3.27 (m, 2H), 2.58−2.51 (m, 4H), 2.47−2.41 (m, 2H), 1.85−1.76 (m, 2H), 1.38−1.11 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 175.4, 171.4, 167.3, 152.9, 62.6, 46.7, 40.3, 34.4, 32.6, 30.0, 23.7, 22.6 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{12}H_{19}N_6O_4$ 311.14623; found 311.14612.

*18-Phenethyl-6,7,12,13,14,15,17,18-octahydro-5H-tetrazolo-[5,1-k][1,4,9,12]oxatriazacyclohexadecene-8,11,16(10H)trione (**11.17**).* The product was obtained as a white solid (29%, 0.120 g, mp 166−168 °C): ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.21 (s, 1H), 7.72 (d, J = 6.0, 1H), 7.31 (d, J = 7.5, 1H), 7.29 (d, J = 4.5, 1H) 7.25−7.23 (m, 1H), 7.07 (d, J = 7.2, 2H), 4.98 (q, J = 7.0, 1H), 4.91 (d, J = 15.5, 1H), 4.28−4.26 (m, 2H), 4.20 (d, J = 15.5, 1H), 3.46−3.36 (m, 1H), 3.17−3.31 (m, 1H), 2.71−2.60 (m, 2H), 2.57−2.36 (m, 6H), 2.33−2.20 (m, 2H), 1.88−1.71 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.9, 171.0, 167.3, 156.3, 139.1, 128.9, 128.2, 126.8, 62.6, 46.6, 43.5, 40.4, 34.8, 34.4, 31.7, 30.2, 23.8, 22.4 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{20}H_{27}N_6O_4$ 415.20883; found 415.20859.

*10-(2-(Methylthio)ethyl)-18-phenethyl-6,7,12,13,14,15,17,18-octahydro-5H-tetrazolo[5,1-k][1,4,9,12]oxatriazacyclohexadecene-8,11,16(10H)-trione (**11.18**).* The product was obtained as a white solid (25%, 0.122g, mp 194−196 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.27−7.25 (m, 2H), 7.18 (t, J = 7.3, 2H), 7.11 (d, J = 7.3, 2H), 7.06 (d, J = 7.4, 1H), 7.01 (t, J = 7.7, 1H), 5.36−5.24 (m, 1H), 4.63−4.57 (m, 1H), 4.41−4.26 (m, 2H), 4.21−4.09 (m, 1H), 3.39−3.36 (m, 1H), 3.35−3.21 (m, 2H), 3.16−3.13 (m,1H), 2.71−2.63 (m, 2H), 2.61−2.54 (m, 2H), 2.52−2.46 (m, 2H), 2.43 (d, J = 6.5, 1H), 2.39−2.28 (m, 6H), 2.27−2.22 (m, 2H), 2.18−2.06 (m, 6H), 2.07−2.04 (s, 3H), 1.97 (s, 1H), 1.82−1.77 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.8, 174.1, 171.8, 171.2, 170.6, 155.9, 155.5, 139.9, 139.5, 128.8, 128.7, 128.4, 128.2, 126.6, 126.5, 72.9, 52.00, 46.7, 46.1, 44.1, 42.4, 42.1, 40.4, 35.2, 35.0, 34.2, 31.8, 31.6, 30.7, 30.5, 30.1, 24.5, 23.2, 22.8, 15.4 ppm; HRMS (ESI/TOF-Q) m/z $[M - H]^+$ calcd for $C_{23}H_{31}N_6O_4S$ 487.2133; found 487.2134.

*10-Isopropyl-6,7,12,13,14,15,17,18-octahydro-5H-tetrazolo-[5,1-k][1,4,9,12]oxatriazacyclohexadecene-8,11,16(10H)-trione (**11.19**).* The product was obtained as a white solid (35%, 0.123 g, mp 208−210 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, DMSO) δ 8.69 (t, J = 5.0, 1H), 8.21 (t, J = 4.8, 1H), 4.82 (d, J = 4.4, 1H), 4.58−4.52 (m, 2H), 4.49−4.47 (m, 1H), 4.44−4.42 (m, 1H), 4.40−4.37 (m, 1H), 3.20−3.09 (m, 2H), 2.59−2.53 (m,1H), 2.50−2.39 (m, 2H), 2.32−2.24 (m,4H), 2.20−2.11(m, 2H), 2.06−1.96 (m,1H), 1.73−1.66 (m,1H), 1.60−1.58 (m, 1H), 0.89 (d, J = 7.0,1H), 0.87 (d, J = 6.9, 3H), 0.83 (d, J = 6.9, 3H), 0.75 (d, J = 6.8, 1H); ^{13}C NMR (126 MHz, DMSO) δ 173.8, 172.2, 168.4, 153.1, 77.6, 45.9, 38.5, 32.3, 32.1, 29.5, 29.4, 23.5, 18.6, 16.8 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{15}H_{25}N_6O_4$ 353.19318; found 353.19324.

*1-Benzyl-18′-phenethyl-6′,7′,12′,13′,14′,15′,17′,18′-octahydrospiro[piperidine-4,10′-tetrazolo[5,1-k][1,4,9,12]-oxatriazacyclohexadecene]-8′,11′,16′(5′H)-trione (**11.20**).* The product was obtained as a white solid (21%, 0.120 g, mp 165−167 °C). A mixture of rotamers was observed and the majority of rotamers taken: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.67 (d, J = 8.5, 1H), 7.45 (d, J = 8.1, 1H), 7.41−7.31 (m, 3H), 7.31−7.23 (m, 3H), 7.19 (td, J = 6.8, 1.3, 1H), 7.13 (d, J = 7.5, 1H), 7.09 (d, J = 7.6, 1H), 5.39−5.31 (m, 1H), 4.47−4.43 (m, 1H), 4.39−4.32 (m, 1H), 4.32−4.22 (m, 1H), 3.77 (s, 1H), 3.53−3.43 (m, 1H), 3.42 (s, 1H), 3.38−3.23 (m, 1H), 3.05−2.97 (m, 1H), 2.78−2.51 (m, 3H), 2.50−2.39 (m, 2H), 2.39−2.31 (m, 4H), 2.30−2.16 (m, 3H), 2.16−2.04 (m, 2H), 2.05−1.87 (m, 2H), 1.80−1.73 (m, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.1, 171.7, 171.5, 155.6, 140.0, 130.2, 128.7, 128.4, 128.3, 126.5, 79.5, 61.2, 47.7, 46.9, 43.0, 42.2, 41.0, 39.8, 35.1, 33.4, 31.9, 31.6, 30.9, 25.1, 24.5 ppm; HRMS (ESI/TOF-Q) m/z $[M - H]^+$ calcd for $C_{31}H_{38}N_7O_4$ 572.29908; found 572.29901.

*5-Phenyl-5,6,11,12,13,14,16,17-octahydrotetrazolo[5,1-k][1,-4,9,12]oxatriazacyclopentadecene-7,10,15(9H)-trione (**11.21**).* The product was obtained as a white solid (20%, 0.074g, mp 159−161 °C): ^{1}H NMR (500 MHz, MeOD) δ 7.81 (d, J = 16.1,1H), 7.65−7.64 (m, 2H), 7.47−7.41 (m, 3H), 6.66 (d, J = 16.0,1H), 4.69 (s, 2H), 3.37 (s, 2H), 2.37−2.23 (m, 4H), 1.89−1.84 (m, 2H), 1.71−1.65 (m, 2H); ^{13}C NMR (126 MHz, MeOD) δ 172.3, 171.4, 155.6, 144.4, 128.8, 127.2, 126.5, 78.8, 60.8, 36.7, 32.1, 25.5, 24.2, 23.3 ppm; HRMS (ESI/TOF-Q) m/z $[M + H]^+$ calcd for $C_{17}H_{21}N_6O_4$ 373.16188; found 373.16174.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.joc.6b01430.

X-ray data for **5.14** (CIF)

X-ray data for **5.17** (CIF)

X-ray data for **11.8** (CIF)

DOI: 10.1021/acs.joc.6b01430
J. Org. Chem. 2016, 81, 8789−8795

NMR spectra and crystal structure determinations (PDF)

AUTHOR INFORMATION

Corresponding Author

*E-mail: a.s.s.domling@rug.nl, www.drugdesign.nl.

Notes

The authors declare no competing financial interest.

ACKNOWLEDGMENTS

The work was financially supported (A.D.) by the NIH (1R01GM097082-01) and by Innovative Medicines Initiative (Grant Agreement No. 115489). The research (K.K., J.K.-T.) was carried out with the equipment purchased thanks to the financial support of the European Regional Development Fund in the framework of the Polish Innovation Economy Operational Program (Contract No. POIG.02.01.00-12-023/08). E.M.M.A. was supported by the Egyptian government. This project has also received funding from the European Union's Horizon 2020 research and innovation programme under MSC ITN "Accelerated Early stage drug dIScovery" (AEGIS), grant agreement No. 675555.

DEDICATION

The authors dedicate this work to Ivar Ugi, the father of modern multicomponent reaction chemistry.

REFERENCES

(1) Suarez-Jimenez, G.-M.; Burgos-Hernandez, A.; Ezquerra-Brauer, J.-M. *Mar. Drugs* **2012**, *10*, 963.

(2) Gu, X.; Wang, L.; Gao, Y.-f.; Ma, W.; Li, Y.-m.; Gong, P. *Tetrahedron: Asymmetry* **2014**, *25*, 1573.

(3) Reza Kazemizadeh, A.; Ramazani, A. *Curr. Org. Chem.* **2012**, *16*, 418.

(4) Fetzer, U.; Ugi, I. *Justus Liebigs Annalen der Chemie* **1962**, *659*, 184.

(5) Xue, D.-Q.; Mollo, E.; Cimino, G.; Guo, Y.-W. *Helv. Chim. Acta* **2009**, *92*, 1428.

(6) Beck, B.; Larbig, G.; Mejat, B.; Magnin-Lachaux, M.; Picard, A.; Herdtweck, E.; Dömling, A. *Org. Lett.* **2003**, *5*, 1047.

(7) Henkel, B.; Beck, B.; Westner, B.; Mejat, B.; Dömling, A. *Tetrahedron Lett.* **2003**, *44*, 8947.

(8) Banfi, L.; Riva, R. The Passerini Reaction. In *Organic Reactions*; John Wiley & Sons, Inc.: New York, 2004.

(9) Andreana, P. R.; Liu, C. C.; Schreiber, S. L. *Org. Lett.* **2004**, *6*, 4231.

(10) Denmark, S. E.; Fan, Y. *J. Am. Chem. Soc.* **2003**, *125*, 7825.

(11) Kusebauch, U.; Beck, B.; Messer, K.; Herdtweck, E.; Dömling, A. *Org. Lett.* **2003**, *5*, 4021.

(12) Wang, S.-X.; Wang, M.-X.; Wang, D.-X.; Zhu, J. *Angew. Chem.* **2008**, *120*, 394.

(13) Barreto, A. d. F. S.; Vercillo, O. E.; Wessjohann, L. A.; Andrade, C. K. Z. *Beilstein J. Org. Chem.* **2014**, *10*, 1017.

(14) Leon, F.; Rivera, D. G.; Wessjohann, L. A. *J. Org. Chem.* **2008**, *73*, 1762−1767.

(15) Heinis, C. *Nat. Chem. Biol.* **2014**, *10*, 696.

(16) Driggers, E. M.; Hale, S. P.; Lee, J.; Terrett, N. K. *Nat. Rev. Drug Discovery* **2008**, *7*, 608.

(17) Hewitt, W. M.; Leung, S. S. F.; Pye, C. R.; Ponkey, A. R.; Bednarek, M.; Jacobson, M. P.; Lokey, R. S. *J. Am. Chem. Soc.* **2015**, *137*, 715.

(18) Whitty, A.; Zhong, M.; Viarengo, L.; Beglov, D.; Hall, D. R.; Vajda, S. *Drug Discovery Today* **2016**, *21*, 712−717.

(19) Valiyaveetil, F. I.; Sekedat, M.; MacKinnon, R.; Muir, T. W. *J. Am. Chem. Soc.* **2006**, *128*, 11591.

(20) Yurek-George, A.; Cecil, A. R. L.; Mo, A. H. K.; Wen, S.; Rogers, H.; Habens, F.; Maeda, S.; Yoshida, M.; Packham, G.; Ganesan, A. *J. Med. Chem.* **2007**, *50*, 5720.

(21) Zarganes-Tzitzikas, T.; Chandgude, A. L.; Dömling, A. *Chem. Rec.* **2015**, *15*, 981.

(22) Liao, G. P.; Abdelraheem, E. M. M.; Neochoritis, C. G.; Kurpiewska, K.; Kalinowska-Tłuścik, J.; McGowan, D. C.; Dömling, A. *Org. Lett.* **2015**, *17*, 4980.

(23) White, C. J.; Yudin, A. K. *Nat. Chem.* **2011**, *3*, 509.

(24) Masson, G.; Neuville, L.; Bughin, C.; Fayol, A.; Zhu, J. *Top. Heterocycl. Chem.* **2010**, *25*, 1.

(25) Wessjohann, L. A.; Rivera, D. G.; Vercillo, O. E. *Chem. Rev.* **2009**, *109*, 796.

(26) Riva, R.; Banfi, L.; Basso, A. The Passerini Reaction. In *Science of Synthesis: Multicomponent Reactions*; Müller, T. J. J., Ed.; Thieme: Stuttgart, 2013; Vol. *1*, pp 327−414.

DOI: 10.1021/acs.joc.6b01430
J. Org. Chem. 2016, 81, 8789−8795