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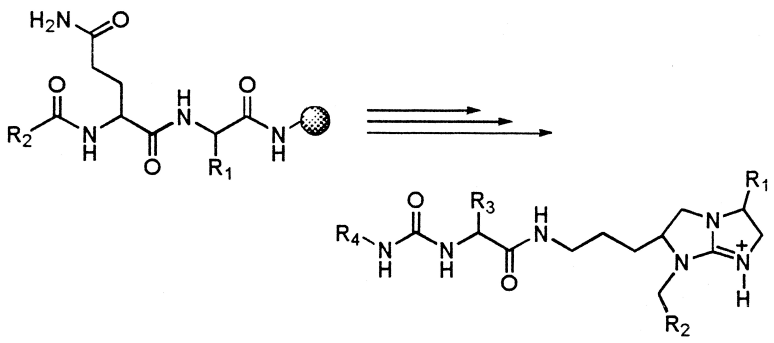

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Tethered Libraries: Solid-Phase Synthesis of Substituted Urea-Linked Bicyclic Guanidines

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The general concept of tethered combinatorial libraries of compounds in which two pharmacophores are found is described. In particular, an improved method for the solid-phase synthesis of bicyclic guanidines from reduced *N*-acylated dipeptides, and its use in the synthesis of urea-linked bicyclic guanidines, is described. The exhaustive reduction of glutamine-containing resin-bound *N*-acylated dipeptides, using borane-THF, generated compounds containing three secondary amines and one primary amine. Following selective trityl protection of the primary amine, treatment of the three secondary amines with thiocarbonyldiimidazole ($CSIm_2$) and mercuric acetate ($Hg(OAc)_2$) generated the resin-bound bicyclic guanidines. Following trityl deprotection, an Fmoc-amino acid was coupled. Upon removal of the Fmoc protecting group, the resulting primary amine was treated with hexyl isocyanate to generate the urea-linked bicyclic guanidines. The desired products were cleaved from the resin using hydrogen fluoride. The selection of building blocks and characterization of controls for the synthesis of a combinatorial library is discussed.

Recently, much attention has been focused on the development of solid-phase methods for the synthesis of small molecules. Due to its inherent advantages, solid-phase synthesis is now used as a technique for the preparation of large numbers of individual compounds, mixtures, and libraries.[1] This has allowed for the synthesis of large numbers of compounds in a short time period, enabling their use in high-throughput screening.[2] In our laboratory, we have concentrated our efforts toward the development of new strategies for the synthesis of individual- and mixture-based libraries of small heterocyclic compounds from amino acids and short peptides.[2] Recently, we have focused on the development of tethered combinatorial libraries in which two or more pharmacophores are present. Guanidino-tethered compounds have exhibited potent antimicrobial activity against *Helicobacter pylori* which has been widely accepted as a major causative factor in peptic ulcer diseases.[3,4]

Guanidines have long been the focus of considerable attention as a ubiquitous moiety incorporated into many drugs with numerous therapeutic applications and biological activities. Reported examples include the antiulcer drug *cimetidine* and the antihypertensive agent *pinacidil*.[3] Other therapeutic applications of guanidines have been as an adrenergic neuron blocking agent;[5] antitumor agent;[6] antihistaminic, antiinflammatory, antidiabetic, and antibacterial drugs;[7] H_2 receptor agonist/antagonist;[8] hypotensive;[9] potassium/ATP channel opener;[10] neuronal Na^+/Ca^{2+} channel blockers;[11] glutamate release inhibitor and antiischemic agent;[12] antiseizure agents;[13] and HIV-1 protease inhibitors.[14] Recent studies from our laboratory showed that trisubstituted bicyclic guanidines[15] exhibit antifungal activity against *Candida albicans* and *Cryptococcus neoformans*.[16] Compounds containing the urea scaffold are found in many biologically active synthetic targets.[17] Some of these targets influence the activities of acid gastric secretion and healing of chronic gastric ulcers in rats,[18] antioxidant derivatives,[19] inhibitors of acyl-CoA: cholesterol O-acyltransferase (ACAT),[20] and metaloprotenase enzyme inhibitors.[21] The urea-linked bicyclic guanidine scaffold also has two unique properties, namely, the guanidino part is basic in nature whereas the urea functionality is neutral. Considering the various novel aspects of these two biologically active moieties, the preparation of urea-linked bicyclic guanidines was carried out.

In a continuation of our efforts to identify highly active compounds from synthetic combinatorial libraries (SCLs),[22] we describe herein an efficient strategy for the solid-phase synthesis of bicyclic guanidines and urea-linked bicyclic guanidine libraries.

Starting from *p*-methylbenzhydrylamine (MBHA) resin, a Boc-amino acid was coupled to the resin. Following deprotection of the *N*-terminal protecting group (Boc) and neutralization, the free amine was coupled with Boc-glutamine. Following Boc deprotection, the resulting primary amine of the resin-bound dipeptide **1** was acylated with a wide variety of available carboxylic acids. Exhaustive reduction of the resulting resin-bound *N*-acylated dipeptide **2** (Scheme 1) using borane−THF generated compound **3** containing three secondary amines and one primary amine.[15,23,24] The primary amine was selectively protected with a trityl group. The resin-bound trityl protected polyamine **4** was treated with thiocarbonyldiimidazole ($CSIm_2$)[15] followed by mercuric acetate ($Hg(OAc)_2$) to yield the resin-bound bicyclic guanidine **5**. Following deprotection of the trityl group and neutralization, an Fmoc-amino acid was coupled in the presence of *N*,*N*′-diisopropylcarbodiimide (DIC) and *N*-hydroxybenzotriazole (HOBt). Following re-

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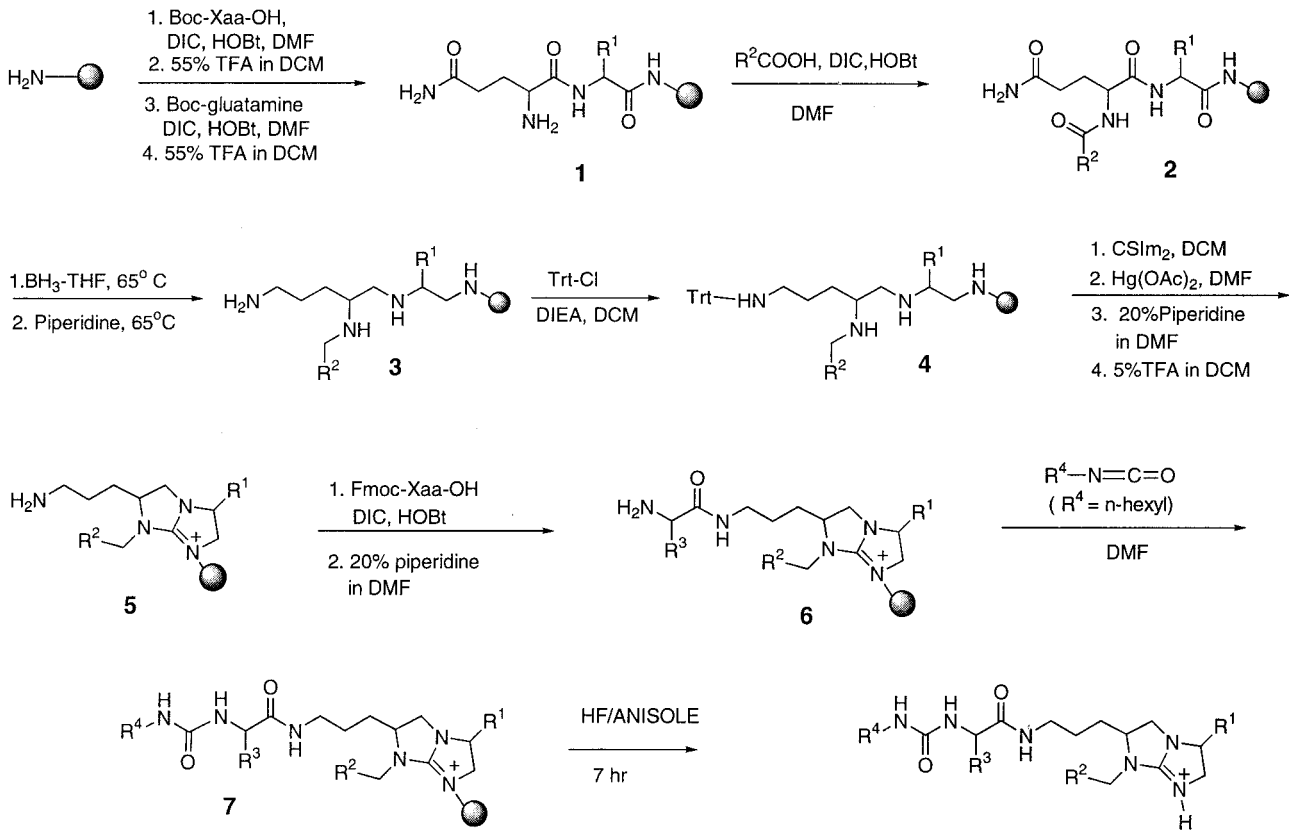
Scheme 1. Solid-Phase Synthesis of Urea-Linked Bicyclic Guanidines



moval of the Fmoc group with piperidine in DMF, the resulting primary amine of **6** was treated with hexyl isocyanate to form the resin-bound urea-linked bicyclic guanidine **7**. Each step (exhaustive reduction of the amide bonds with borane−THF, bicyclic guanidine cyclization, and the hexyl isocyanate treatment) was carried out under an anhydrous nitrogen atmosphere.

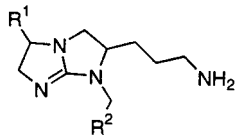
The possibility of racemization during either the exhaustive reduction of the resin-bound *N*-acylated dipeptides using borane−THF, selective protection of primary amine by tritylation, cyclization to form bicyclic guanidines, or the hexyl isocyanate reaction was monitored using reverse-phase high-performance liquid chromatography (RP-HPLC). Different diastereomeric pairs of glutamine-containing *N*-acylated dipeptide that were found not to coelute were synthesized as controls for the chemical reactions involved. Negligible racemization (<1%) was observed, and this has also been described in our previous reports.[15] Following deprotection of the trityl group after selective tritylation of the primary amine of the reduced polyamine (i.e., before cyclization), RP-HPLC data indicated that negligible (<1%) racemization occurred during tritylation in the presence of excess base. Comparison of RP-HPLC data of two different diastereomer pairs of **5** indicated that negligible or no racemization (<1%) occurred during cyclization on treatment with $CSIm_2$ and $Hg(OAc)_2$. Similar results were observed following treatment with hexyl isocyanate.

Following optimization of the different steps, we expanded the individual controls by separately varying the substituent at each of these three positions. Compounds representing more than 40 amino acids for the first and third position of diversities (R^1, R^3), and 48 carboxylic acids for the second position of diversity (R^2), were synthesized.

Amino acids that generate a reactive functionality after reduction (for example, asparagine and glutamine) were not included in the R^1 position. Also, those amino acids having an extra amine functionality, such as lysine, were not used for position R^1 or R^3. We observed that *N*-benzyl derivatives resulting from reduced substituted benzoic acids for the second position of diversity (R^2) are cleaved during the required 7 h HF cleavage. These carboxylic acids were excluded from the library synthesis. Menthoxy acetic acid was included for the second position of diversity (R^2) in the libraries, as the individual control compound obtained was found to yield the hydroxy ethyl functionality, in good purity and yield, upon complete cleavage of menthol moiety following 7 h exposure to HF (**5j** and **7j**). In most cases, substituted phenylacetic acid derivatives were used for N-acylation at the second position of diversity (R^2).

Although previous syntheses of bicyclic guanidines[15] achieved reasonable results, it was found that for more highly substituted bicyclic guanidines the cyclization of the resin-bound tritylated polyamines **4** was frequently incomplete when using only $CSIm_2$. A side product, having a molecular weight of 34 mass units higher than the mass of the desired bicyclic guanidine, was formed corresponding to the cyclic thiourea derivative intermediate. This problem was alleviated successfully by treatment of the intermediate with $Hg(OAc)_2$. The resin-bound trityl protected polyamine was first treated with $CSIm_2$ once. The uncyclized cyclic thiourea formed after treatment with $CSIm_2$ was then driven to completion using $Hg(OAc)_2$ in DMF. This afforded compound **5** (Scheme 1**)**

Table 1. Synthesis of Bicyclic Guanidines **5**[a,b]



entry	R^1	R^2	MW (calcd)	MW (found)
5a	$-CH_2C_6H_5$	$-CH_2C_6H_5$	362.5	363.6 (MH^+)
5b	$-CH_2C_6H_5$	$-CH_2CH(C_6H_5)_2$	452.6	453.8 (MH^+)
5c	$-C_6H_{11}$	$-CH_2C_6H_5$	354.5	355.7 (MH^+)
5d	$-CH_2CH(CH_3)_2$	$-CH_2C_6H_5$	328.5	329.6 (MH^+)
5e	$-CH_2C_6H_5$	$-CH_2C_6H_5$	362.5	363.4 (MH^+)
5f	$-CH(CH_3)_2$	$-CH_2C_6H_5$	314.5	315.5 (MH^+)
5g	$-CH_2C_6H_5$	$-CH(C_2H_5)CH_2CH_3$	342.5	343.7 (MH^+)
5h	$-CH_2C_6H_5$	$-CH_2CH_2C_6H_5$	376.5	377.4 (MH^+)
5i	$-CH_2C_6H_5$	$-C_6H_{11}$	354.5	355.5 (MH^+)
5j	$-CH_2C_6H_5$	$-CH_2OH$	302.4	303.2 (MH^+)

[a] Yields were >70% by mass. The percent yield was calculated according to the theoretical loading of the resin at 1.0 mequiv/g. [b] Compounds **5e** and **5f** are of *R* configuration and all others are of *S* configuration for the first position of diversity (R^1).

without any trace of cyclic thiourea derivative or a negligible amount of uncyclized material as determined by LC-MS of **5** (Table 1). Ten randomly selected compounds of **5** out of 75 using 35 Boc-AA's (R^1) and 40 carboxylic acids (R^2) (listed in the Supporting Information) are presented in Table 1. Furthermore, the appearance of a peak at ~157−158 ppm (**7a**−**c**) in the ^{13}C NMR confirmed the presence of a guanidino moiety.

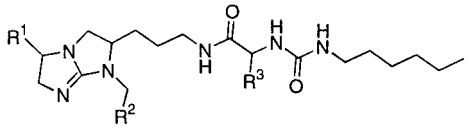
After successful completion of the cyclization to generate the resin-bound bicyclic guanidines, the trityl group was removed from the primary amine. Fmoc amino acids were coupled to the primary amine of the resin-bound bicyclic guanidines using the classical reagents DIC and HOBt, followed by deprotection of the Fmoc group to generate intermediate **6**. Treatment of intermediate **6** with hexyl isocyanate (R^4 = *n*-hexyl) in DMF generated **7** (Table 2). The appearance of a peak at ~162−164 ppm (**7a**−**c**) in ^{13}C NMR confirmed the presence of an urea moiety. All the above individual controls obtained were found to be in reasonable yield (see Table 2) and purity with negligible racemization.

Only building blocks that produced cyclized compounds having purities higher than 80% were considered for inclusion in the synthesis of libraries. Following the synthesis of individual controls, we selected 35 different amino acids for the first position (R^1) of diversity, 40 carboxylic acids for the second position (R^2), and 34 different amino acids for the third position of diversity (R^3) for synthesis of positional scanning combinatorial libraries (SCLs).[25] There is one defined position for each position of diversity. Predetermined isokinetic ratios[23] for each protected amino acid (both Boc and Fmoc) and carboxylic acid were used during coupling of mixtures. R^4 was left constant (hexyl isocyanate) in order to obtain a positional scanning synthetic combinatorial library (PS-SCL)[25] since isokinetic ratios are not known for the use of isocyanates. This single position defined SCL was then screened to explore the most active compounds identifying the individual functionality at each position of diversity. The preparation of this positional scanning combinatorial library containing 47 600 (35 R^1 × 40 R^2 × 34 R^3) different urea-linked bicyclic guanidines and its screening in different assays for the identification of highly active compounds will be reported elsewhere.

Summary

The general concept of tethered combinatorial libraries was described. In particular, an improved solid-phase synthesis of bicyclic guanidines and the subsequent derivatization to urea-linked bicyclic guanidines was presented. We found that the reaction of the thiourea derivative intermediate, formed as the side product during cyclization of *N*-acylated reduced dipeptides, was driven to completion to generate the bicyclic guanidine by treatment with mercuric acetate. Modified dipeptides have been successfully exploited for the preparation of positional scanning urea-linked bicyclic guanidine libraries using the "libraries from libraries" approach.[22] The

Table 2. Synthesis of Urea-Linked Bicyclic Guanidines **7**[a]



entry	R^1	R^2	R^3	MW (calcd)	MW (found)	yield (%)[b]	purity[c]
7a	$-CH_2C_6H_5$	$-CH_2C_6H_5$	$-CH_2C_6H_5$	636.9	637.6 (MH^+)	69	81
7b	$-CH_2C_6H_5$	$-CH_2CH(C_6H_5)_2$	$-CH_2C_6H_5$	727.0	727.6 (MH^+)	77	85
7c	$-C_6H_{11}$	$-CH_2C_6H_5$	$-CH_2C_6H_5$	628.9	629.4 (MH^+)	66	82
7d	$-CH_2CH(CH_3)_2$	$-CH_2C_6H_5$	$-CH_2C_6H_5$	602.9	603.7 (MH^+)	76	84
7e	$-CH_2C_6H_5$	$-CH_2C_6H_5$	$-CH_2C_6H_5$	636.9	637.8 (MH^+)	61	80
7f	$-CH(CH_3)_2$	$-CH_2C_6H_5$	$-CH_2C_6H_5$	588.8	589.7 (MH^+)	82	80
7g	$-CH_2C_6H_5$	$-CH(C_2H_5)CH_2CH_3$	$-CH_2C_6H_5$	616.9	617.7 (MH^+)	62	81
7h	$-CH_2C_6H_5$	$-CH_2CH_2C_6H_5$	$-CH_2C_6H_5$	650.9	651.8 (MH^+)	82	81
7i	$-CH_2C_6H_5$	$-C_6H_{11}$	$-CH_2C_6H_5$	628.9	629.7 (MH^+)	60	82
7j	$-CH_2C_6H_5$	$-CH_2OH$	$-CH_2C_6H_5$	576.8	577.5 (MH^+)	90	79
7k	$-CH_2C_6H_5$	$-CH_2C_6H_5$	-H	546.8	547.4 (MH^+)	88	82

[a] Compounds **7e** and **7f** are of *R* configuration and all others are of *S* configuration for the first position of diversity (R^1). All the amino acids for the third position of diversity (R^3) are of *S* configuration except **7k**. [b] The percent yield was calculated according to the theoretical loading of the resin at 1.0 mequiv/g. [c] The purity was calculated from the relative peak areas of the HPLC chromatograms.

discovery of novel active compounds of this scaffold will be reported elsewhere.

Experimental Section

MBHA resin, 1% divinylbenzene, 100−200 mesh, 1 mequiv/g substitution, and *N*,*N*′-diisopropylcarbodiimide (DIC) were purchased from Chem Impex Intl. (Wood Dale, IL). Boc, Fmoc-amino acid derivatives, and *N*-hydroxybenzotriazole (HOBt) were purchased from Calbiochem-Novabiochem Corp. (San Diego, CA) and Bachem Bioscience Inc. (Philadelphia, PA). Trifluoroacetic acid (TFA) and HF were purchased from Halocarbon (River Edge, NJ) and Air Products (San Marcos, CA), respectively. Triphenylmethyl chloride (Trt-Cl) was purchased from Acros (San Diego, CA). All other reagents and anhydrous solvents were purchased from Aldrich Chemical Co. (Milwaukee, WI). Analytical RP-HPLC was performed on a Beckman System Gold Instrument (Fullerton, CA). Samples were analyzed using a Vydac 218TP54 C18 column (0.46 × 25 cm). LC-MS (APCI) were recorded on a Finnigan Mat LCQ mass spectrometer (ThermoQuest Corporation, CA) at 214 nm using a Betasil C18, 3 μm, 100Å, 3 × 50 mm column.

Typical Procedure for the Individual Synthesis of Urea-Linked Bicyclic Guanidines. A polypropylene mesh packet was sealed with 100 mg of *p*-methylbenzhydrylamine (MBHA) resin (1 mequiv/g, 100−200 mesh).[26] Reactions were carried out in polypropylene bottles. The resin was washed with dichloromethane (DCM) followed by neutralization with 5% diisopropylethylamine (DIEA) in DCM and washed with DCM. The first Boc-amino acid (6 equiv) was coupled using DIC and HOBt (6 equiv each) in anhydrous DMF for 2 h. Following washes with DMF (6 times), Boc deprotection was performed using 55% TFA in DCM for 30 min, followed by washing with DCM (2 times), 2-propanol (IPA) (2 times), and DCM (2 times). Following neutralization, coupling of Boc-glutamine, deprotection of the Boc group, and neutralization were performed in the same manner as described above. The above dipeptides were then acylated with a carboxylic acid (10 equiv) in the presence of DIC and HOBt (10 equiv each) in anhydrous DMF overnight. Completeness of the coupling was verified by the ninhydrin test.[27]

(1) Exhaustive Reduction of Amide Groups by BH_3−THF. The exhaustive reduction of the N-acylated dipeptide was carried out in 50 mL glass conical tubes under nitrogen. To each tube was added the resin packet (0.1 mequiv resin, 100 mg of starting resin, 0.40 mequiv carbonyl) and boric acid (12×, 297 mg). Trimethyl borate (0.53 mL, 12×) was added followed by the slow addition of 16 mL of borane−THF complex (1 M, 40×). After cessation of hydrogen evolution, the capped tubes were heated at 65 °C for 72 h in a heating block followed by decantation of the reaction solution and quenching with MeOH. The resin packet was then washed with DMF and methanol (MeOH) (4 times). The resin was treated with piperidine at 65 °C for 20 h to disproportionate the borane complexes.[15] Following decantation of the piperidine−borane solution, the resin packet was washed with DMF (4 times), DCM (4 times), and MeOH (2 times) and dried.

(2) Cyclization To Form the Bicyclic Guanidine. The resin was neutralized with 5% DIEA in DCM followed by tritylation of the primary amine using Trt-Cl (10 equiv) in the presence of DIEA (25 equiv) for 2 h. The coupling of Trt-Cl was repeated overnight to ensure completeness. Following neutralization, the resin was dried under vacuum. Cyclization was performed in two steps: (a) the trityl protected reduced N-acylated dipeptides were treated with thiocarbonyldiimidazole ($CSIm_2$) (24 equiv) in anhydrous DCM (0.37 M) overnight under nitrogen (inside a glovebox) followed by washes with anhydrous DCM (5 times), IPA (2 times), and DCM (4 times) and (b) following neutralization, the resin was dried under vacuum and was treated overnight with mercuric acetate ($Hg(OAc)_2$) (10 equiv) in anhydrous DMF followed by washes in DMF (6 times) and DCM (3 times). The resin was treated with 20% piperidine in DMF for 1 h to dissociate the mercuric salts, followed by washes in DMF (4 times) and DCM (3 times).

(3) Coupling of Fmoc-Amino Acid. The trityl group was deprotected using 5% TFA in DCM (3 times × 10 min), and the resin packet was washed and neutralized with 5% DIEA in DCM. An Fmoc-amino acid was coupled using the same procedure described above. The Fmoc group was removed with 20% piperidine in DMF for 30 min. The resin was then washed with DMF (4 times) and DCM (3 times).

(4) Formation of Urea. The resin was treated with 1% TFA in DCM (10 min, 2 times) followed by neutralization and then was treated with hexyl isocyanate (1.5 equiv) in anhydrous DMF under nitrogen for 2 h to yield the ureas. The resin was cleaved by anhydrous HF in the presence of anisole at 0 °C for 7 h,[28] and the cleaved product was extracted with 95% acetic acid in H_2O and lyophilized. The identity of the compounds was determined by using LC-MS, HRMS, ^{1}H NMR, and ^{13}C NMR data.

***N*-{3-[5-Benzyl-1-(2-phenylethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)-carbonyl]phenylalaninamide (7a).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.84 (t, J = 6.8 Hz, 3H), 1.18−1.31 (m, 10H), 1.44 (t, J = 4.7 Hz, 1H), 1.55−1.56 (m, 1H), 2.73−2.78 (m, 2H), 2.84−3.02 (m, 8H), 3.07−3.10 (m, 1H), 3.25 (t, J = 9.1 Hz, 1H), 3.35−3.37 (m, 1H), 3.53−3.55 (m, 1H), 3.63 (t, J = 9.1 Hz, 1H), 3.90 (t, J = 9.4 Hz, 1H), 3.99 (m, 1H), 4.23 (m, 1H), 4.32−4.33 (m, 1H), 6.02 (t, J = 5.6 Hz, 1H), 7.15−7.35 (m, 15H), 7.96 (t, J = 5.6 Hz, 1H), 9.32 (s, 1H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 13.89, 22.06, 23.83, 25.99, 27.93, 29.85, 31.0, 32.94, 37.1, 37.92, 38.93, 39.00, 43.91, 48.14, 52.98, 54.49, 59.32, 64.22, 126.13, 126.62, 126.82, 127.96, 128.39, 128.59, 128.91, 129.19, 129.26, 136.56, 137.64, 137.91, 157.41, 163.34, 172.0. HRMS (FAB) *m*/*z* 637.4253 found ($[M + H]^+$), 637.4230 calculated for $C_{39}H_{53}N_6O_2$ ($[M + H]^+$).

***N*-{3-[5-Benzyl-1-(3,3-diphenylpropyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)carbonyl]phenylalaninamide (7b).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.84 (t, J = 6.8 Hz, 3H), 1.18−1.40 (m, 11H), 2.28−2.31 (m, 1H), 2.34−2.37 (m, 1H), 2.71−2.75 (dd, J = 7.9 Hz, J = 13.6 Hz, 1H), 2.82−2.98 (m, 9H), 3.02−3.04 (m, 1H), 3.11−3.13 (m, 1H), 3.28−3.32 (m, 1H), 3.60 (t, J = 9.2 Hz, 1H), 3.84−3.85 (m, 1H), 3.91−

3.97 (m, 2H), 4.15−4.16 (m, 1H), 4.32−4.33 (m, 1H), 6.0−6.03 (dd, J = 6.1 Hz, J = 11.2 Hz, 1H), 7.15−7.36 (m, 20H), 7.93 (t, J = 5.5 Hz, 1H), 9.14 (s, 1H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 13.88, 22.05, 23.69, 25.98, 27.65, 29.85, 30.98, 31.71, 37.19, 38.00, 41.70, 47.84, 48.60, 52.90, 54.41, 59.23, 63.74, 126.11, 126.32, 126.82, 127.32, 127.44, 127.94, 128.48, 128.52, 128.57, 129.19, 136.54, 137.86, 144.01, 144.41, 157.37, 162.95, 171.88. HRMS (FAB) *m*/*z* 727.4683 found ([M + H]$^+$), 727.4699 calculated for $C_{46}H_{59}N_6O_2$ ([M + H]$^+$).

***N*-{3-[5-Cyclohexyl-1-(2-phenylethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)-carbonyl]phenylalaninamide (7c).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.84 (t, J = 6.7 Hz, 3H), 0.94−0.98 (m, 2H), 1.13−1.34 (m, 10H), 1.45−1.70 (m, 8H), 2.69−2.74 (dd, J = 8.1 Hz, J = 13.5 Hz, 2H), 2.79−2.92 (m, 3H), 2.99−3.01 (m, 1H), 3.09−3.11 (m, 1H), 3.32−3.43 (m, 5H), 3.48−3.51 (m, 2H), 3.53−3.55 (m, 2H), 3.67 (t, J = 9.7 Hz, 1H), 3.90 (t, J = 9.5 Hz, 1H), 4.26−4.31 (m, 2H), 5.98−6.01 (dd, J = 6.6 Hz, J = 11.0 Hz, 1H), 7.14−7.33 (m, 10H), 7.94 (t, J = 5.5 Hz, 1H), 9.25 (s, 1H). ^{13}C NMR (125 MHz, DMSO-d_6) δ 13.89, 22.05, 23.84, 25.24, 25.31, 25.59, 25.98, 27.49, 28.21, 28.31, 29.85, 31.00, 32.88, 37.86, 38.88, 39.00, 43.98, 49.50, 51.03, 54.44, 63.42, 64.20, 126.10, 126.61, 127.93, 128.38, 128.91, 129.16, 137.65, 137.89, 157.37, 163.87, 171.97. HRMS (FAB) *m*/*z* 629.4550 found ([M + H]$^+$), 629.4543 calculated for $C_{38}H_{57}N_6O_2$ ([M + H]$^+$).

***N*-[(Hexylamino)carbonyl]-*N*-{3-[5-isobutyl-1-(2-phenylethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}phenylalaninamide (7d).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.83−0.89 (m, 9H), 1.19−1.37 (m, 11H), 1.44−1.45 (m, 1H), 1.55−1.62 (m, 3H), 2.72−2.74 (m, 2H), 2.80 (m, 4H), 2.84−2.93 (m, 1H), 2.99−3.01 (m, 1H), 3.08−3.11 (m, 1H), 3.27−3.29 (dd, J = 3.5 Hz, J = 8.9 Hz, 1H), 3.37−3.43 (m, 2H), 3.53−3.60 (m, 1H), 3.70−3.73 (dd, J = 5.3 Hz, J = 8.0 Hz, 1H), 4.01−4.02 (m, 1H), 4.26−4.31 (m, 2H), 6.00−6.02 (d, J = 7.8 Hz, 1H), 7.15−7.32 (m, 10H), 7.94 (t, J = 5.4 Hz, 1H), 9.20 (s, 1H).

***N*-{3-[(5R)-5-Benzyl-1-(2-phenylethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)carbonyl]phenylalaninamide (7e).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.82−0.86 (m, 3H), 1.15−1.35 (m, 11H), 1.66−1.75 (m, 1H), 2.70−2.75 (m, 2H), 2.79−2.99 (m, 9H), 3.05−3.08 (m, 1H), 3.38−3.41 (m, 2H), 3.50−3.57 (m, 1H), 3.85−4.01 (m, 2H), 4.30−4.31 (m, 2H), 5.99−6.00 (d, J = 8.5 Hz, 1H), 7.14−7.36 (m, 15H), 7.94 (t, J = 5.6 Hz, 1H), 9.22 (s, 1H).

***N*-[(Hexylamino)carbonyl]-*N*-{3-[(5R)-5-isopropyl-1-(2-phenylethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}phenylalaninamide (7f).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.81−0.89 (m, 9H), 1.19−1.37 (m, 11H), 1.76−1.83 (m, 2H), 2.71−2.79 (m, 2H), 2.81−3.03 (m, 6H), 3.09−3.11 (m, 1H), 3.41−3.58 (m, 3H), 3.70 (t, J = 8.2 Hz, 1H), 3.93 (t, J = 7.7 Hz, 1H), 4.26−4.34 (m, 3H), 6.01−6.02 (d, J = 7.6 Hz, 1H), 7.15−7.31 (m, 10H), 7.96 (t, J = 5.6 Hz, 1H), 9.22 (s, 1H).

***N*-{3-[5-Benzyl-1-(2-ethylbutyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)carbonyl]phenylalaninamide (7g).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.80−0.86 (m, 9H), 1.14−1.34 (m, 14H), 1.43−1.45 (m, 1H), 1.54−1.58 (m, 2H), 2.70−2.75 (dd, J = 8.2 Hz, J = 13.6 Hz, 1H), 2.85−3.11 (m, 7H), 3.15−3.20 (dd, J = 9.5 Hz, J = 14.4 Hz, 1H), 3.33 (t, J = 9.1 Hz, 1H), 3.64 (t, J = 9.4 Hz, 3H), 3.97 (t, J = 9.3 Hz, 1H), 4.09 (t, J = 6.8 Hz, 1H), 4.21 (t, J = 8.2 Hz, 1H), 4.30−4.33 (m, 1H), 5.99−6.01 (d, J = 8.2 Hz, 1H), 7.16−7.36 (m, 10H), 7.97 (t, J = 5.6 Hz, 1H), 9.20 (s, 1H).

***N*-{3-[5-Benzyl-1-(3-phenylpropyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)-carbonyl]phenylalaninamide (7h).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.84 (t, J = 6.8 Hz, 3H), 1.19−1.30 (m, 7H), 1.41−1.43 (m, 3H), 1.49−1.52 (m, 2H), 1.79−1.86 (m, 2H), 2.54−2.58 (dd, J = 5.9 Hz, J = 10.4 Hz, 2H), 2.71−2.76 (dd, J = 8.0 Hz, J = 13.6 Hz, 1H), 2.85−3.08 (m, 6H), 3.14−3.23 (m, 2H), 3.34−3.37 (m, 1H), 3.62−3.67 (m, 3H), 3.98−4.00 (m, 2H), 4.22 (m, 1H), 4.32−4.33 (m, 1H), 6.03 (t, J = 3.9 Hz, 1H), 7.15−7.35 (m, 15H), 7.96 (t, J = 5.4 Hz, 1H), 9.34 (s, 1H).

***N*-{3-[5-Benzyl-1-(cyclohexylmethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)-carbonyl]phenylalaninamide (7i).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.82−0.90 (m, 3H), 1.10−1.34 (m, 12H), 1.44 (t, J = 4.8 Hz, 2H), 1.57−1.67 (m, 9H), 2.71−2.75 (dd, J = 8.1 Hz, J = 13.6 Hz, 1H), 2.85−3.12 (m, 8H), 3.34 (t, J = 8.9 Hz, 1H), 3.61−3.65 (m, 3H), 3.96 (t, J = 9.2 Hz, 1H), 4.7−4.10 (m, 1H), 4.19−4.22 (m, 1H), 4.29−4.32 (m, 1H), 6.00−6.01 (d, J = 7.9 Hz, 1H), 7.16−7.35 (m, 10H), 7.96 (t, J = 5.6 Hz, 1H), 9.17(s, 1H).

***N*-{3-[5-Benzyl-1-(2-hydroxyethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*-[(hexylamino)-carbonyl]phenylalaninamide (7j).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.84 (t, J = 6.7 Hz, 3H), 1.19−1.33 (m, 12H), 1.46 (t, J = 4.5 Hz, 1H), 1.53−1.56 (dd, J = 5.2 Hz, J = 9.8 Hz, 1H), 2.73−2.75 (m, 1H), 2.85−3.06 (m, 8H), 3.18−3.27 (m, 1H), 3.30−3.40 (m, 1H), 3.46−3.57 (m, 1H), 3.63 (t, J = 9.1 Hz, 2H), 3.96 (m, 1H), 4.05 (m, 1H), 4.31 (m, 2H), 6.00−6.02 (d, J = 7.9 Hz, 1H), 7.16−7.36 (m, 10H), 7.95 (t, J = 5.5 Hz, 1H), 9.07 (s, 1H).

***N*^{1}-{3-[5-Benzyl-1-(2-phenylethyl)-2,3,5,6-tetrahydro-1*H*-imidazo[1,2-*a*]imidazol-2-yl]propyl}-*N*^{2}-[(hexylamino)-carbonyl]glycinamide (7k).** ^{1}H NMR (500 MHz, DMSO-d_6): δ 0.84 (t, J = 6.6 Hz, 3H), 1.22−1.37 (m, 10H), 1.51−1.53 (d, J = 9.5 Hz, 1H), 1.63 (m, 1H), 2.77−2.80 (m, 2H), 2.84−2.97 (m, 3H), 3.02−3.11 (m, 3H), 3.25 (t, J = 8.9 Hz, 2H), 3.37−3.64 (m, 4H), 3.89−3.91 (m, 1H), 3.99 (m, 1H), 4.26 (m, 1H), 6.02 (s, 1H), 6.12 (s, 1H), 7.22−7.35 (m, 10H), 7.82 (t, J = 5.6 Hz, 1H), 9.20 (s, 1H).

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Supporting Information Available. LC-MS of individual bicyclic guanidines and urea-linked bicyclic guanidines, and HRMS and NMR spectra (both ^{1}H and ^{13}C) of some

selected urea-linked bicyclic guanidines. This material is available free of charge via the Internet at http://pubs.acs.org.

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