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Synthetic Methods |*Hot Paper*|

α-Amino Acid-Isosteric α-Amino Tetrazoles

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Abstract: The synthesis of all 20 common natural proteinogenic and 4 otherα-amino acid-isosteric α-amino tetrazoles has been accomplished, whereby the carboxyl group is replaced by the isosteric 5-tetrazolyl group. The short process involves the use of the key Ugi tetrazole reaction followed by deprotection chemistries. The tetrazole group is bioisosteric to the carboxylic acid and is widely used in medicinal chemistry and drug design. Surprisingly, several of the common α-amino acid-isosteric α-amino tetrazoles are unknown up to now. Therefore a rapid synthetic access to this compound class and non-natural derivatives is of high interest to advance the field.

Introduction

α-Amino acids are among the most important known classes of organic compounds, with multiple applications in nutrition, medicine, materials, chemistry, and biochemistry.[1] Sometimes, however, the physicochemical properties of α-amino acids are not suitable for a specific application and isosteric derivatives can be more suitable. For example some α-amino acids have a very high metabolic turnover or a low solubility at a specific pH. Well established isosteres of the carboxylic acid are hydroxamic, phosphonic and sulfonic acids, sulfonamides, acylsulfonamides, and tetrazoles, to name just a few.[2] A specialized bioisostere database mentions >2000 COOH bioisosteres.[3] Among COOH isosteres, the tetrazole group is of special interest since it has a comparable pKa, a similar size, a similar spatial arrangement of the heteroatom lone pairs, and a similar molecular electrostatic potential, and therefore often undergoes a very similar receptor–ligand interaction (Figure 1).[4] However, the tetrazole group often exhibits a prolonged half-life, owing to enhanced metabolic stability,[5] enhanced spatial delocalization of the negative charge, and better membrane penetration resulting from increased lipophilicity.[6] Therefore, tetrazole represents the first-choice bioisosteric group if the corresponding COOH has issues in medicinal chemistry projects.[7]

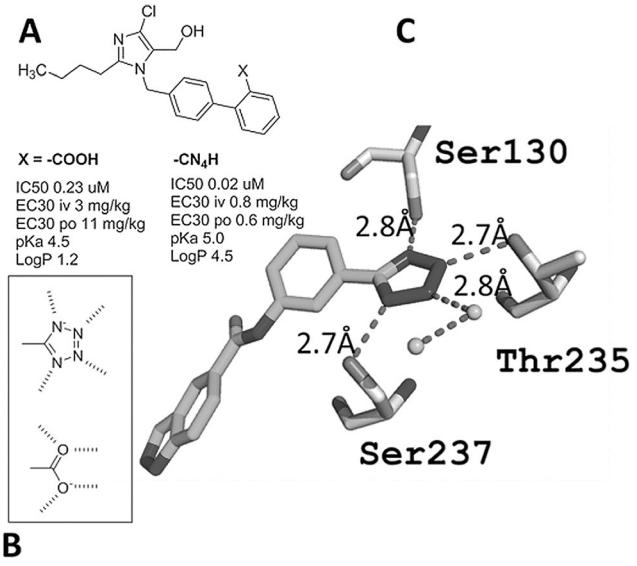

Figure 1. A) The bioisosteric carboxylic acid and tetrazolyl moieties in two Losartan derivatives;[8] B) the four lone pairs of the four nitrogen atoms of tetrazole and the two oxygen atoms of the carboxylic acid point into similar regions in space; C) receptor ligand interactions around a tetrazolate moiety in a β-lactamase (PDB ID:4DE1) showing the involvement of all four nitrogen lone pairs.[9]

In 1959, McManus and Herbst first reported two independent methods to prepare several α-amino acid tetrazole analogues.[10] Later, in 2002, Demko and Sharpless disclosed a novel [3+2] cycloaddition of azide onto nitrile or nitrilium to synthesize several endogenous α-amino acid-bioisosteric α-amino tetrazoles.[11] However, these approaches possess several disadvantages, such as high reaction temperatures, use of toxic reagents, metal catalyst utilization, and failure to synthesize all 20 natural proteinogenic α-amino acid-bioisosteric tetrazoles (Scheme 1).

To our knowledge, the preparation of all 20 natural proteinogenic α-amino acid-bioisosteric tetrazoles by one synthetic strategy has not been reported to date. Multicomponent reac-

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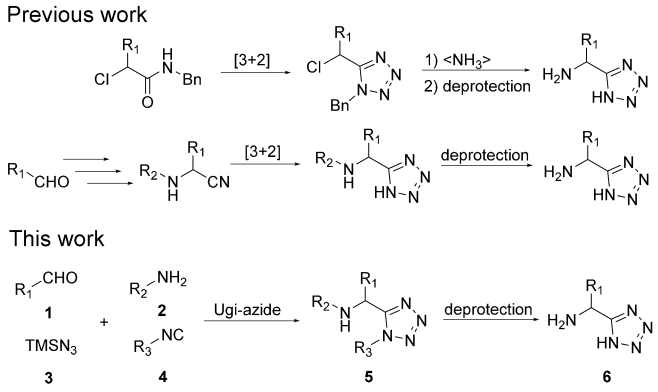
tions (MCRs) are a powerful way to afford the desired target compounds with great structural diversity in a short reaction sequences.[12] Isocyanide-based multicomponent reactions (IMCRs) are of special interest, owing to the variety of starting materials available and a plethora of transformations reported.[13] Ugi and Meyr reported the first application of IMCRs to synthesize tetrazole derivatives in 1961.[14] They employed NaN_3 to replace the carboxylic acid in the 3-component Passerini reaction to give 1,5-disubstituted α-hydroxymethyl tetrazole derivatives. Afterwards, numerous tetrazole-fused structure-varied derivatives were synthesized by IMCRs to yield libraries of screening compounds applied in high-throughput screening.[15]

In this study, we instead employed the IMCR Ugi tetrazole synthesis with suitable starting materials aldehyde **1**, cleavable amine **2**, and isocyanide **4** in the presence of the azide source $TMSN_3$ **3** to access α-amino acid-bioisosteric α-amino tetrazoles **6** in a convergent way and to synthesize, for the first time, all the 20 endogenous α-amino acids isosteres (Scheme 1).



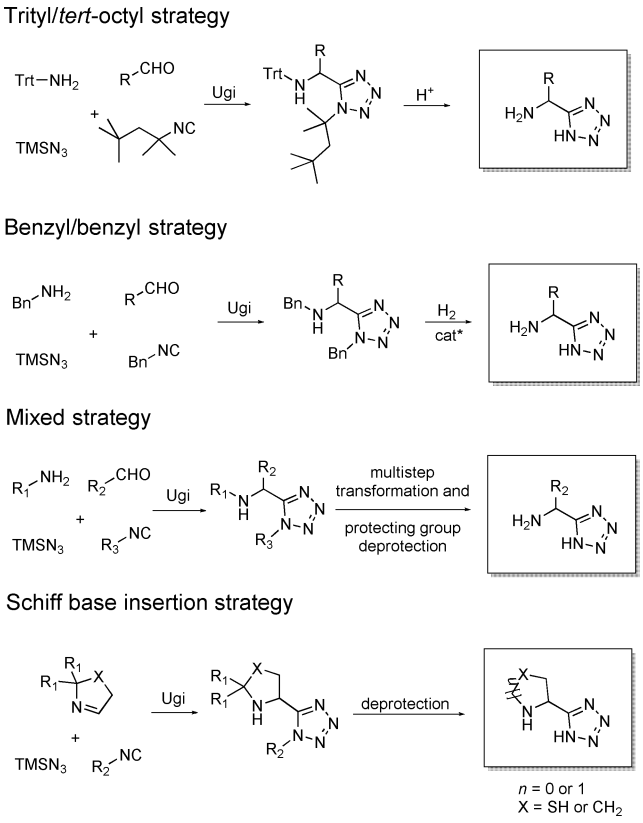
Scheme 1. Known syntheses of α-amino acid-bioisosteric α-amino tetrazoles and our approach.

Although the Ugi tetrazole synthesis is well established and has found multiple applications in the synthesis of different scaffolds, there has, to our knowledge, been no report on the use of this reaction to afford α-amino acid-bioisosteric α-amino tetrazoles. Therefore, we describe herein the synthesis of all 20 natural proteinogenic and 4 other amino acid analogues through the $TMSN_3$ IMCR process.

Results and Discussion

In this study, we aimed to establish a novel method based on IMCRs to conveniently synthesize α-amino acid isosteric tetrazoles. Four protecting-group strategies were devised to account for compatibility with the amino acid side chains (Scheme 2): trityl/*tert*-octyl strategy, benzyl/benzyl strategy, mixed strategy, and Schiff base insertion strategy.

It is well established that ammonia is a poor amine component in most IMCRs, leading to multiple side products and overall poor yields.[16] Therefore an ammonia surrogate must

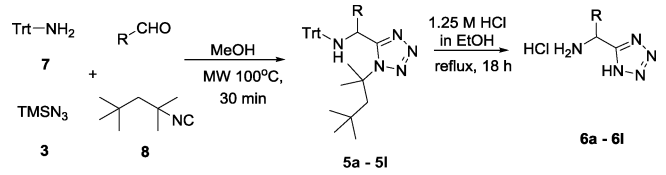


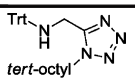
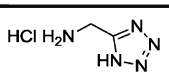
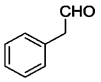
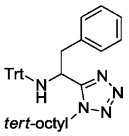
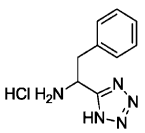
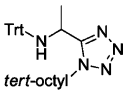
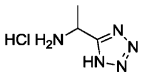
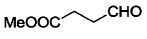
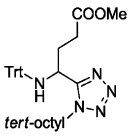
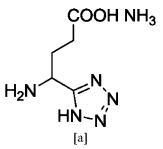
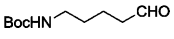
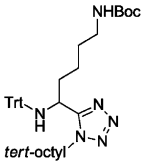
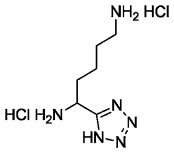
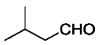
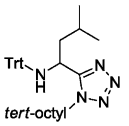
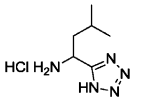
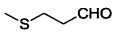
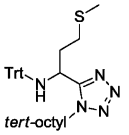
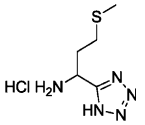
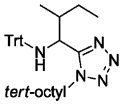
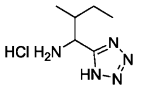
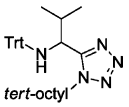
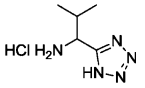
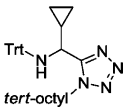
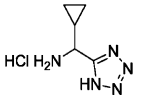
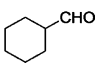
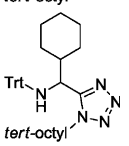
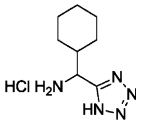
Scheme 2. Four different protecting-group strategies to target α-amino acid-bioisosteric α-amino tetrazoles.

be used in our MCR strategy. Based on our previous work, tritylamine **7** could be used as such in the Ugi reaction, giving moderate to high yields after mild cleavage of the trityl protecting group.[17] The combination of **7** and Walborsky's isocyanide (*tert*-octylisocyanide) **8** allowed for simultaneous deprotection of the Ugi products to give the α-amino tetrazoles in just one simple step (Scheme 2).

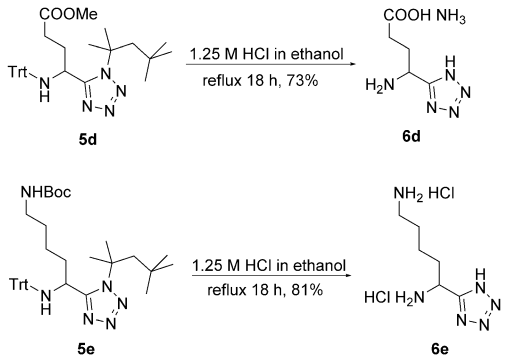
We therefore mixed the commercially available aldehydes and tritylamine **7** in the presence of methanol and heated in a microwave at 100 °C for 15 min to obtain the Schiff base and then added $TMSN_3$ **3** and *tert*-octyl isocyanides **8**, again under microwave irradiation, to yield the Ugi products containing simple aliphatic or aromatic side chains (**5a**–**l**). Notably, the high reaction temperature was not necessary for the Ugi ring closure but to force the Schiff base formation, as reported previously.[17] The final Ugi tetrazoles **5a**–**l** were isolated in moderate to excellent yields by column chromatography on silica (Table 1). Next, the trityl and *tert*-octyl groups were simultaneously removed in ethanolic HCl (1.25 M) by heating at reflux overnight to give the α-amino acid-isosteric tetrazoles **6a**–**l** in moderate to excellent yields (Table 1). For the amino acids Glu and Lys, the aldehydes are not commercially available and had to be synthesized. Aldehyde **10** for Glu Ugi product **5d** could be conveniently synthesized from the commercial mixed acid chloride methyl ester **9** (Scheme 4).[18] Glu analogue **6d** could be obtained by simultaneous removal of the trityl group and the *tert*-octyl group, as well as hydrolysis of the methyl ester

Table 1. Trityl/*tert*-octyl strategy. Yields of the Ugi products (**5a–l**) and α-amino acid tetrazoles (**6a–l**).



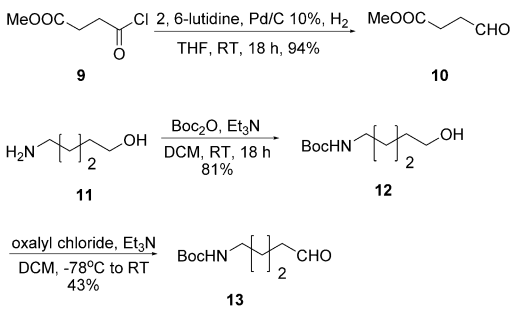
Entry	Aldehyde	Ugi product **5**	Yield [%]	α-amino acid tetrazole **6**	Yield [%]
a	HCHO		56		51
b			57		35
c	MeCHO		58		61
d			36	 [a]	73
e			96		81
f			62		82
g			44		99
h			49		71
i			48		63
j			52		82
k			54		70

by heating the Ugi product **5d** at reflux in ethanolic 1.25 M HCl (Scheme 3). Aldehyde **13**, for the Lys Ugi product **5e**, was synthesized by *N*-Boc protection of



Scheme 3. Simultaneous deprotection of Ugi products **5d** and **5e**.

commercial alcohol **11**, followed by Swern oxidation at −78 °C (Scheme 4).[19] Lys analogue **6e** could be obtained by simultaneous removal of the Boc, trityl and *tert*-octyl groups by heating the Ugi product **5e**

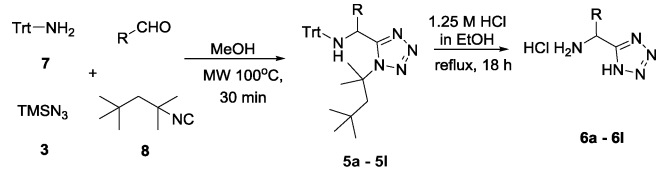


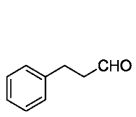
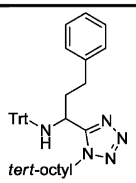
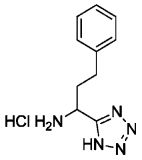
Scheme 4. Synthesis of aldehydes **10** and **13** for Ugi products **5d** (Glu) and **5e** (Lys).

at reflux in ethanolic 1.25 M HCl (Scheme 3). Racemic and commercial 2-methylbutanal led to the Ile Ugi product **5h** and the Ile analogue **6h** as a mixture of two diastereomers in 1:1 ratio (Table 1, **5h** and **6h**). This lack of stereoselectivity is a well-established feature of Ugi reactions.[20]

For side-chain-functionalized amino acid derivatives, the trityl/*tert*-octyl strategy did not work because of the lack of Schiff base formation, presumably due to the harsh reaction conditions required for trityl-Schiff base formation. Therefore, in several cases, a benzyl/benzyl strategy was employed. In contrast to the trityl/*tert*-octyl strategy, the benzyl amine **14** and benzylisocyanide **15** were used, which reacted smoothly with the aldehydes and $TMSN_3$ **3** at room temperature to give the Ugi tetrazoles in

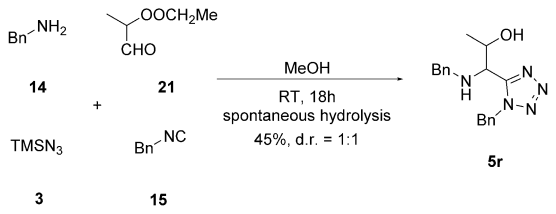
Table 1. (Continued)



Entry	Aldehyde	Ugi product **5**	Yield [%]	α-amino acid tetrazole **6**	Yield [%]
l			53		99

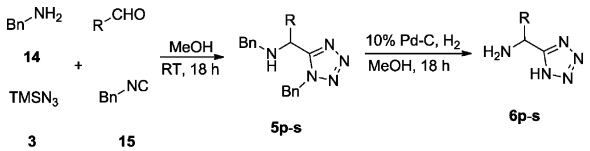
[a] Purification was carried out on a Dowex 50 cation-exchange column eluted with 2N aqueous NH_3.

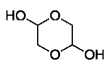
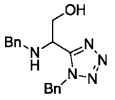
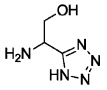
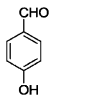
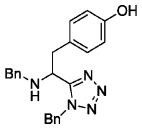
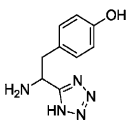
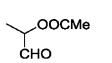
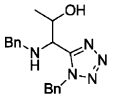
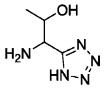
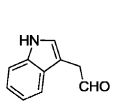
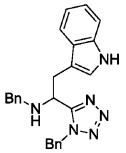
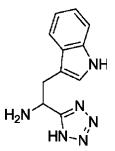
moderate to excellent yields (Table 2). Notably, for the Thr isostere **5r**, a simultaneous ester hydrolysis took place during the Ugi reaction (Scheme 5). Similar to Ile precursor **5h**, the racemic aldehyde **21** led to the Thr precursor **5r** and Ile analogue **6r** as a mixture of two diastereomers in 1:1 ratio (Table 2, **5r** and **6r**). For the analogous Tyr, Thr, and Trp Ugi products, the aldehydes are also not commercially available and had to be synthesized. Commercial **16** was first esterified in refluxing methanolic HCl, yielding methylester **17**; subsequent reduction with $LiAlH_4$ and then Parikh–Doering oxidation were performed to give aldehyde **18**.[21] Aldehyde **21**, for the Thr Ugi product **5q**, was synthesized by using a previously reported procedure.[22] Firstly, 4,4-dimethoxybutan-2-one **19** was reduced by $NaBH_4$ at low temperature. The obtained alcohol was acetylated by acetic anhydride in the presence of trimethylamine to form acetyl acetal **20**. Subsequently, compound **20** was deprotected by using cation-exchange resin Dowex 50 to give 4-oxobutan-2-yl acetate **21**. Aldehyde **24** was prepared by a similar method to aldehyde **18**. Commercial acetic acid **22** was first reduced to alcohol **23** by using $LiAlH_4$ in THF followed by Parikh–Doering oxidation (Scheme 6). It is important to note that aldehydes **18**, **21**, and **24** are unstable and cannot be stored and should therefore be used immediately after synthesis.



Scheme 5. The Ugi reaction and an unexpected spontaneous hydrolysis to form Thr Ugi product **5r**.

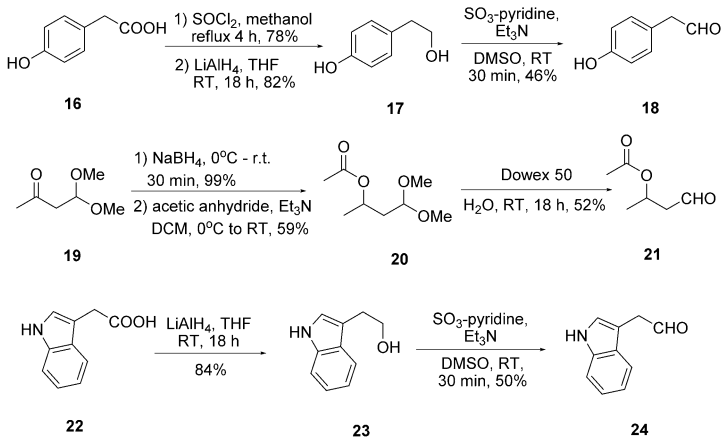
Table 2. Benzyl/benzyl strategy. Yields of the Ugi products (**5p–s**) and α-amino acid tetrazoles (**6p–s**).



Entry	Aldehyde	Ugi product **5**	Yield [%]	α-Amino acid tetrazole **6**[a]	Yield [%]
p			69		52
q			20		66
r			45		45
s			28		57

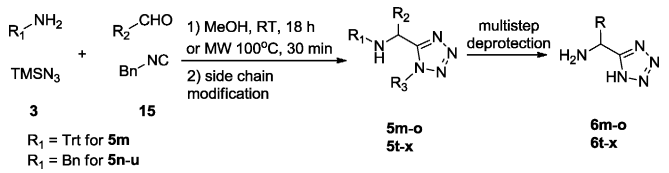
[a] Purification was carried out on a Dowex 50 cation-exchange column eluted with 2N aqueous NH_3.

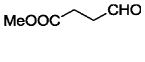
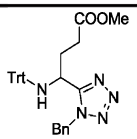
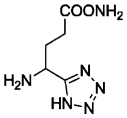
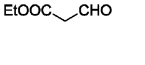
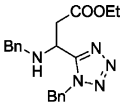
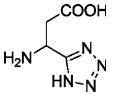
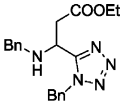
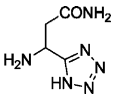
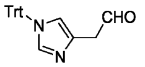
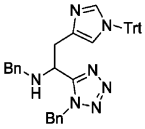
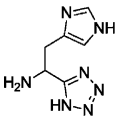
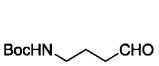
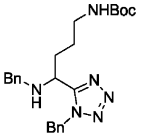
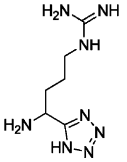
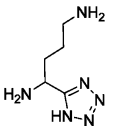
For the other side-chain-functionalized amino derivatives (**5m**, **5n**, **5t**, **5u**, and **5x**), a mixed strategy was performed (Table 3). The trityl/*tert*-octyl strategy could not be employed for the Gln Ugi product **5m** because the amide side chain is destroyed upon refluxing in an acidic solution required for the *tert*-octyl cleavage. Moreover, the benzyl/benzyl strategy was also not suitable, owing to the spontaneous formation of γ-lactam **5m″** (Scheme 7). Therefore we applied a trityl/benzyl strategy to produce Gln Ugi product **5m**. The bulky tritylamine group in **5m** avoided the spontaneous γ-lactamization that occurred for the benzyl analogue **5m″**. To afford Gln precursor **5m′**, the methyl ester in Ugi product **5m** was hydrolyzed by LiOH. The formation of amide **5m′** was accomplished by a coupling step with 1-hydroxybenzotriazole (HOBt), 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDCI), *N*,*N*-diisopropylethylamine (DIEA), and $(NH_4)_2CO_3$ as the ammonia source.[23] The deprotection of amide **5m′** to yield Gln analogue **6m** was carried out in two more steps; trityl removal under mild acidic condition with trifluoroacetic acid (TFA) in CH_2Cl_2 at room temperature and hydrogenolytic benzyl deprotection by H_2 and Pd on charcoal. Aldehyde **10**, for Gln, was also used for Glu (Scheme 4). Similarly the tritylamine strategy is not compatible with Asp or Asn Ugi products. Although the Schiff base **25** was clearly formed, the subsequent Ugi reaction did not proceed,



Scheme 6. The preparation of aldehydes **18**, **21** and **24** for Ugi products **5q** (Tyr), **5r** (Thr), and **5s** (Trp).

Table 3. Mixed strategy: Yields of the Ugi products (**5m–o** and **5t–x**) and α-amino acid tetrazoles (**6m–o** and **6t–x**).

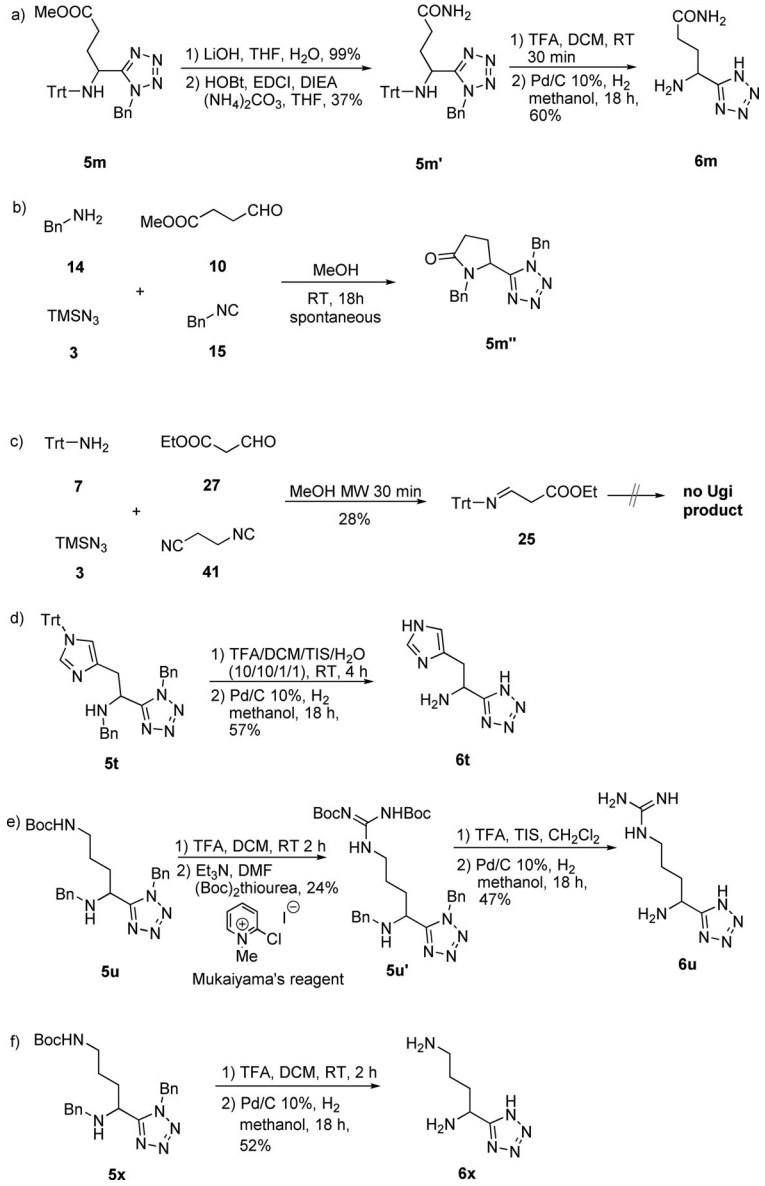


Entry	Aldehyde	Ugi product **5**	Yield [%]	α-Amino acid tetrazole **6**[a]	Yield [%]
m			45		52
n			60		66
o			60		45
t			28		57
u			54		47
x					52

[a] Purification was carried out on a Dowex 50 cation-exchange column eluted with 2N aqueous NH_3.

even under prolonged reaction times, presumably due to the bulkiness and enolic character of **25** (Scheme 7) blocking the attack of the isocyanide **41**. Alternatively, we employed the benzyl/benzyl strategy to prepare the Ugi product **5n**. Deprotection to **6n** was accomplished by first ester hydrolysis using aqueous LiOH, followed by double debenzylation with H_2 and Pd on charcoal. Notably, the benzyl/benzyl strategy also facilitated the transformation of carboxylic acid to amide without any loss of amide under the mild deprotection conditions for benzyl groups (Scheme 7). Likewise, for amide **5o** the same coupling reaction as for amide **5m′** was carried out (Scheme 7). For the His Ugi product **5t**, the trityl-protected imidazole aldehyde **30** had to be synthesized starting from commercial imidazole acetic acid **28**, by a sequence of esterification, tritylation, and reduction by using diisobutylaluminium hydride (DIBAL) at low temperature (Scheme 8).[24] Aldehyde **30** was used in the Ugi reaction directly without further purification to afford the Ugi product **5t**. His analogue **6t** was obtained by TFA-mediated hydrolysis using triisopropylsilane (ITS) as a cation scavenger in medium yield (57%; Scheme 7).[25] For Arg Ugi product **5u**, a more complicated synthetic strategy was required based on the demanding guanidine side chain. We utilized the benzyl/benzyl strategy to obtain Ugi product **5u**. Before forming the guanidine group, the tailed NH_2 group was deprotected under mild acidic conditions and was neutralized to the free amine. Next, di-*tert*-butyl thiourea with Mukaiyama's reagent as a coupling reagent was added in the presence of triethylamine to form di-Boc-protected guanidine-containing tetrazole derivative **5u′** (Scheme 7).[26] Di-*tert*-butyl thiourea **32** was prepared as described previously.[27] The Boc groups in the guanidine moiety in **5u′** were removed by using triisopropylsilane as a cation scavenger with TFA, followed by hydrogenolytical benzyl group removal leading to Arg analogue **6u** in good yield (47%, two steps). Ornithine is another non-proteinogenic endogenous α-amino acid of great importance, being the metabolic product of L-arginine formed by the enzyme arginase. Thus we also synthesized Orn analogue **6x** by simply removing the Boc group and benzyl groups of the Ugi product **5x** (Scheme 7). The non-commercial aldehyde **35**, for **5x**, was prepared by using a procedure similar to that for aldehyde **13** (Scheme 8).

For the amino acid analogues of Pro and Cys, yet another strategy had to be devised. For the Cys analogue, the harsh reaction conditions in the trityl/*tert*-octyl strategy could not afford the desired Ugi product either with mercaptoacetaldehyde dimer **39** or with chloroacetaldehyde **42** (Scheme 10). By using the benzyl/benzyl strategy, **42** worked well in the Ugi reaction and **43** could be isolated in 63% yield.



Scheme 7. a) Synthesis of compound **5m**′ and multistep deprotection to afford **6m**; b) unexpected spontaneous cyclization to afford compound **5m**′′ by using the benzyl/benzyl strategy; c) Schiff base **25** formed by using tritylamine as an amino source; d) deprotection of **5t** to give His analogue **6t**; e) insertion of guanidine group to lead to compound **5u**′ and deprotection to afford Arg analogue **6u**; f) deprotection of **5x** to give Orn analogue **6x**.

From there, we envisioned the introduction of the mercapto side chain by nucleophilic substitution with NaSH or $NaSOCCH_3$. However, the attempted deprotection of the two Bn groups in the presence of H_2 and Pd/C was not successful. Thus we turned to a different strategy and used a Ugi-ready protected form of mercaptoacetaldehyde: 2,2-dimethylthiazolidine **40**. Thiazolidine **40** is a bench-stable liquid, which is well known to work as a component in the classical Ugi-4CR.[28] Moreover, it can be readily synthesized in good yields in one pot by another MCR, the Asinger-3CR of acetone, ammonia and mercaptoacetaldehyde dimer **39** (Scheme 9).[28,29] The simple reaction of Walborsky's isocyanide **8** with $TMSN_3$ and **40** yielded **5w** in moderate yields. Heating the Ugi product **5w** at reflux in ethanolic HCl provided the Cys analogue **6w** in good yield by N,S-acetal and tetrazole deprotection (Scheme 10 and Table 4). For the Pro analogue, pyrrolidine **36** was oxidized to the cyclic Schiff base Δ1-pyrroline **37**, which exists in equilibrium between cyclo trimer **38**, oligomers, and the monomer (Scheme 9). The oxidation was accomplished with sodium persulfate with catalytic amounts of $AgNO_3$ in water (Scheme 9).[30] Surprisingly, simply mixing Schiff base Δ1-pyrroline **37** with $TMSN_3$ **3** and β-cyanoethylisocyanide **41** in methanol did not lead to the Ugi product **5v**. However upon addition of the Lewis acid $ZnCl_2$ the Ugi tetrazole **5v** was formed in good yields from cleavable β-cyanoethylisocyanide **41** (Scheme 10 and Table 4).

It is worth noting here that the Ugi products generally crystallized well and can be used to study the solid-state conformation and intermolecular contacts, as shown for three typical highly crowded examples in Figure 2.[23]

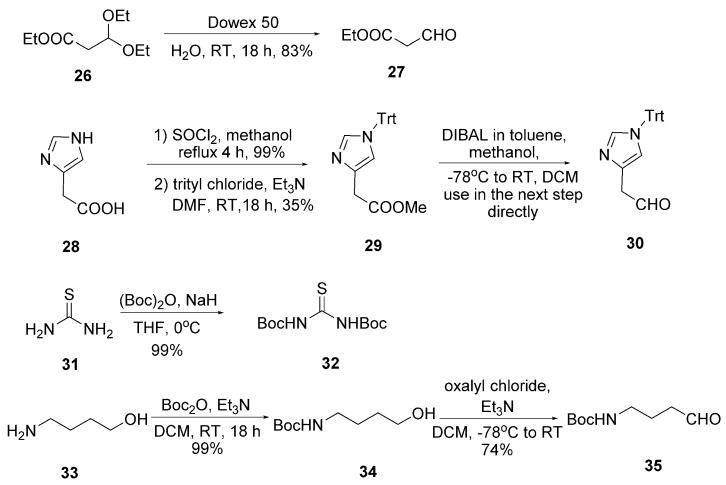
Conclusions

In summary, we have developed a concise and rapid synthetic route to α-amino acid-bioisosteric α-amino tetrazoles, exemplified in the synthesis of all 20 natural proteinogenic amino acids and 4 others by using a key azido-Ugi reaction. To our knowledge, this is the first report of the synthesis of all endogenous natural proteinogenic α-amino acid-bioisosteric α-amino tetrazoles by one general synthetic pathway. Surprisingly, the tetrazole derivatives of the α-amino acids Thr (**6r**), His (**6t**), Arg (**6u**), and Cys (**6w**) are, to our knowledge, hitherto unknown and reported herein for the first time. All compounds were synthesized as racemic mixtures and **6h** and **6r** as diastereomeric mixtures. Investigation of the biological activity of the endogenous natural proteinogenic α-amino acid-bioisosteric α-amino tetrazoles including applications in drug design are ongoing and will be reported in due course.

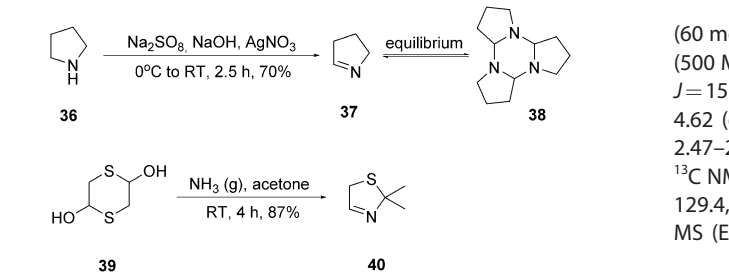
Experimental Section

General methods

Nuclear magnetic resonance spectra were recorded on a Bruker Advance 500 spectrometer [^{1}H NMR (500 MHz), ^{13}C NMR (126 MHz)], a Varian Inova 400 MHz spectrometer [^{1}H NMR (400 MHz), ^{13}C NMR (100 MHz)] or a Varian VXR 300 MHz spectrometer [^{1}H NMR (300 MHz), ^{13}C NMR (75 MHz)]. Chemical shifts δ for ^{1}H NMR were reported in ppm and coupling constants were in Hz. The following abbreviations were used for spin multiplicity: s = singlet, d = doublet, t = triplet, dd = double doublet, m = multiplet. Chemical shifts δ for ^{13}C NMR reported in ppm relative to the sol-



Scheme 8. Synthesis of aldehydes **27**, **30**, and **35** for Ugi products **5 n**, **5 s** and **5 u** and preparation of di-*tert*-butyl sulfinyldicarbamate **32**.



Scheme 9. Synthesis of Schiff bases **37** and **40** for Ugi products **6 v** and **6 w**.

vent peak. Mass spectra (HRMS) were recorded on an Orbitrap XL (Thermo Fisher Scientific; ESI pos. mode, resolution of 60 000 *m/z* 400). Electrospray ionization mass spectra (ESI-MS) were recorded on a Waters Investigator Semi-prep 15 SFC-MS instrument. Mass spectra for deprotected α-amino acid-isosteric α-amino tetrazoles were measured on an API 3000 triple-quadrupole mass spectrometer (Applied Biosystems/MDS Sciex) via a TurbolonSpray source. Thin-layer chromatography was performed on precoated silica gel plates (TLC Silica gel 60 F_{254}). Flash chromatography was performed on a Teledyne ISCO Combiflash R_f, using RediSep R_f Normal-phase Silica Flash Columns (Silica Gel 60 Å, 230–400 mesh). Reagents were available from commercial suppliers and used without any purification unless otherwise noted.

Synthetic procedure following Ugi reaction of trityl/*tert*-octyl and trityl/benzyl strategy

Aldehyde (1 mmol) and tritylamine (1 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 by using microwave irradiation. Isocyanide (1 mmol) and azidotrimethylsilane (1 mmol) were then added into the reaction mixture and further heated at 100 °C for 15 min by using microwave irradiation. The solvent was removed under reduced pressure and the residue was purified by using flash column chromatography.

1,1,1-Triphenyl-*N*-((1-(2,4,4-trimethylpentan-2-yl)-1*H*-tetrazol-5-yl)methyl)methanamine (5 a): The product was obtained as a white solid (254 mg, 56 %). R_f = 0.43 (Petroleum ether:EtOAc 3:1); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 7.56–7.54 (m, 6 H), 7.32–7.29 (m, 6 H), 7.26–7.20 (m, 3 H), 3.73 (dd, *J* = 7.3, 2 H), 1.66 (d, *J* = 1.4 Hz, 2 H), 1.60 (d, *J* = 1.4 Hz, 6 H), 0.65 ppm (d, *J* = 1.5 Hz, 9 H); ^{13}C NMR (126 MHz, $CDCl_3$): δ = 153.8, 145.1, 128.8, 128.3, 127.0, 71.4, 64.3, 53.0, 40.3, 31.7, 30.7, 30.1 ppm; MS (ESI) *m/z* calculated $[M+Na]^+$: 476.28; found $[M+Na]^+$: 476.38.

Synthetic procedure following Ugi reaction of benzyl/benzyl strategy

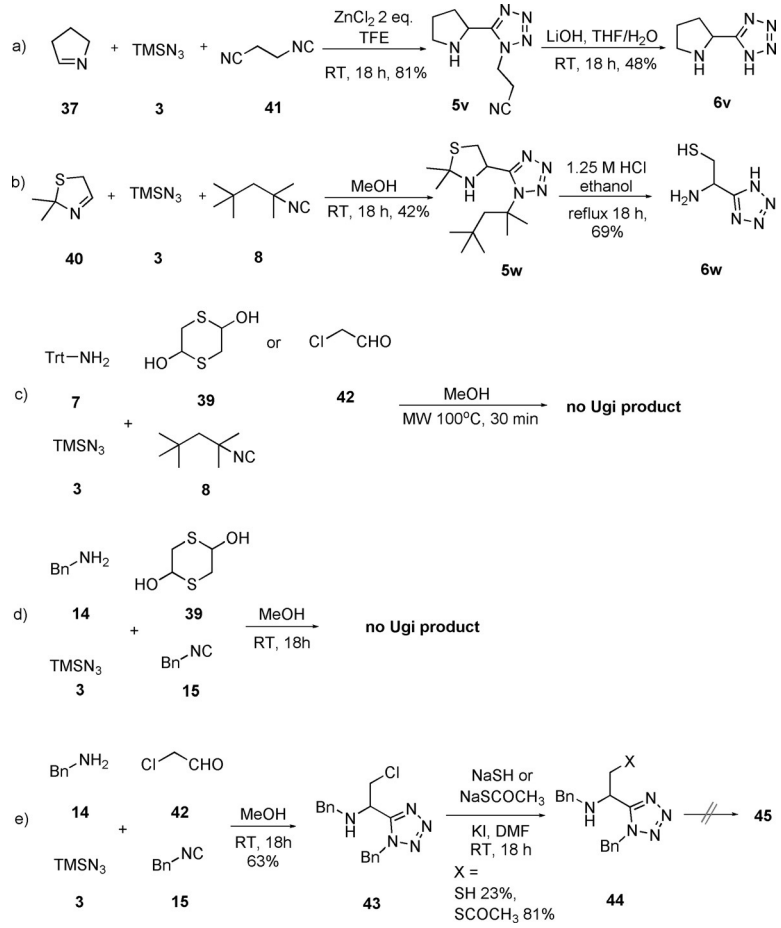
Aldehyde (1 mmol) and benzylamine (1 mmol) were mixed in MeOH (1 mL) in a vial with a magnetic stirring bar. The reaction mixture stirred for 5 min at room temperature. Isocyanide (1 mmol) and azidotrimethylsilane (1 mmol) were then added to the reaction mixture and it was stirred overnight at room temperature. The solvent was removed under reduced pressure and the residue was purified by using flash column chromatography to obtain the desired product.

1-Benzyl-5-(1-benzyl-1*H*-tetrazol-5-yl)pyrrolidin-2-one (5 m″): The product was obtained as a colorless solid (60 mg, 18 %). R_f = 0.49 (Petroleum ether:EtOAc = 2:1); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 7.38–7.24 (m, 6 H), 6.97–6.95 (m, 4 H), 5.40 (d, *J* = 15.5 Hz, 1 H), 5.04 (d, *J* = 14.9 Hz, 1 H), 4.78 (d, *J* = 15.6 Hz, 1 H), 4.62 (dd, *J* = 8.6, 5.0 Hz, 1 H), 3.72–3.61 (m, 1 H), 2.68–2.58 (m, 1 H), 2.47–2.41 (m, 1 H), 2.07–1.97 (m, 1 H), 1.74–1.67 ppm (m, 1 H); ^{13}C NMR (126 MHz, $CDCl_3$): δ = 174.6, 154.5, 135.1, 133.0, 129.5, 129.4, 129.2, 128.5, 128.3, 127.4, 51.1, 50.9, 45.3, 29.5, 24.4 ppm; MS (ESI) *m/z* calculated $[M+Na]^+$: 356.15; found $[M+Na]^+$: 356.30.

Synthetic procedure following Ugi reaction of mixed strategy

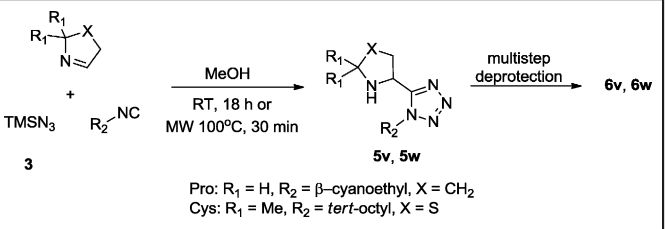
Methyl 4-(1-benzyl-1*H*-tetrazol-5-yl)-4-(tritylamino)butanoate (5 m): The product was obtained as a light yellow or colorless solid (239 mg, 45 %). R_f = 0.32 (Petroleum ether:EtOAc 3:1); ^{1}H NMR (500 MHz, $CDCl_3$): δ = 7.38–7.23 (m, 10 H), 7.22–7.10 (m, 8 H), 7.08–7.01 (m, 2 H), 5.21 (d, *J* = 15.3 Hz, 1 H), 4.78 (d, *J* = 15.3 Hz, 1 H), 4.18 (d, *J* = 5.6 Hz, 1 H), 3.58 (s, 3 H), 2.23–2.17 (m, 1 H), 2.04–1.98 (m, 1 H), 1.92–1.87 (m, 1 H), 1.59–1.52 ppm (m, 1 H); ^{13}C NMR (126 MHz, $CDCl_3$): δ = 173.4, 157.1, 145.2, 133.2, 129.2, 129.0, 128.9, 128.5, 128.1, 127.9, 126.8, 71.4, 51.8, 50.9, 47.0, 31.5, 28.9 ppm; MS (ESI) *m/z* calculated $[M+Na]^+$: 540.24; found $[M+Na]^+$: 540.21.

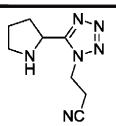
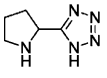
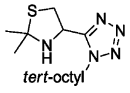
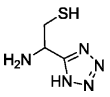
4-(1-Benzyl-1*H*-tetrazol-5-yl)-4-(tritylamino)butanamide (5 m′): Methyl 4-(1-benzyl-1*H*-tetrazol-5-yl)-4-(tritylamino)butanoate (**5 m**; 0.2 g, 0.5 mmol) was dissolved in THF/H_2O = 1/1 (2 mL) containing lithium hydroxide (48 mg, 1 mmol). The reaction mixture was stirred overnight at room temperature. The solvent was removed and the reaction mixture was acidified to pH 3 with 2 M HCl aqueous solution. Then it was extracted with CH_2Cl_2 (10 mL × 3). The organic layer was collected and concentrated to give the product as a white solid with a quantitative yield. It was used directly in the next step without further purification. 4-(1-benzyl-1*H*-tetrazol-5-yl)-4-(tritylamino)butanoic acid (0.2 g, 0.5 mmol) was dissolved in THF (4 mL) and the resultant solution was degassed with N_2. Then DIEA (0.3 mL, 2.3 mmol), HOBt (67 mg, 0.55 mmol), and EDCI (88 μL, 0.055 mmol) were added. The reaction mixture was stirred for 10 min at room temperature. Ammonium carbonate (173 mg, 1.8 mmol) was added. The reaction mixture was further stirred overnight at room temperature. The solvent was removed and CH_2Cl_2 (7 mL) and water (7 mL) were added. The aqueous phase was separated and extracted with CH_2Cl_2 (20 mL × 3). The organic



Scheme 10. a) Synthesis of Ugi products **5v** and **5w**; b) final deprotection to produce **6v** and **6w**; c,d) failure to use trityl/*tert*-octyl strategy or benzyl/benzyl strategy in the preparation of Cys Ugi product; e) another route to obtain the precursor **44** for Cys analogue **6w**.

Table 4. Yields of the Ugi products **5v** and **5w** and α-amino acid tetrazoles **6v** and **6w**.



Entry	Schiff base	Ugi products **5**	Yield [%]	α-Amino acid tetrazole **6**[a]	Yield [%]
v			81		48
w			42		74

[a] Purification was carried out on a Dowex 50 cation-exchange column eluted with 2N aqueous NH_3.

phases were combined and purified with a combiflash machine to give the product as a colorless solid (82 mg, 37%). $R_f = 0.49$ (EtOAc); ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 7.39–7.25$ (m, 9H), 7.23–7.12 (m, 9H), 7.08–7.06 (m, 2H), 5.28 (d, $J = 15.2$ Hz, 1H), 4.80 (d, $J = 15.3$ Hz, 1H), 4.19–4.16 (m, 1H), 2.23–2.17 (m, 1H), 2.02–1.98 (m, 1H), 1.92–1.87 (m, 1H), 1.59–1.54 ppm (m, 1H); ^{13}C NMR (126 MHz, $CDCl_3$): $\delta = 174.1$, 157.0, 145.2, 133.5, 129.2, 129.0, 128.6, 128.2, 128.1, 126.9, 71.7, 51.0, 47.5, 31.5, 30.1 ppm; MS (ESI) *m/z* calculated $[M+Na]^+$: 525.24; found $[M+Na]^+$: 525.41.

Synthetic procedure following Ugi reaction of Schiff base insertion strategy

3-(5-(Pyrrolidin-2-yl)-1*H*-tetrazol-1-yl)propanenitrile (5v): 3,4-Dihydro-2*H*-pyrrole (**37**; 70 mg, 1 mmol) was dissolved in trifluoroethanol (1 mL) and zinc chloride (273 mg, 2 mmol) was added. The reaction mixture stirred for 5 min at room temperature. Then β-cyanoethylisocyanide (**41**; 81 mg, 1 mmol) was added. The reaction mixture was further stirred for 45 min at room temperature. Then 10 drops of aqueous ammonia solution (35%) was added. The reaction mixture was then stirred overnight at room temperature. The reaction was monitored by thin layer chromatography (TLC). When the reaction was completed, the reaction mixture was concentrated under reduced pressure and the residue was purified by using flash column chromatography. The product was a yellow solid (156 mg, 81%). $R_f = 0.42$ (CH_2Cl_2:MeOH 8:2); ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 5.03–4.94$ (m, 1H), 4.85–4.78 (m, 1H), 4.70–4.64 (m, 1H), 3.15–3.06 (m, 3H), 3.01–2.92 (m, 1H), 2.37–2.22 (m, 2H), 2.04–1.84 ppm (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$): $\delta = 157.1$,

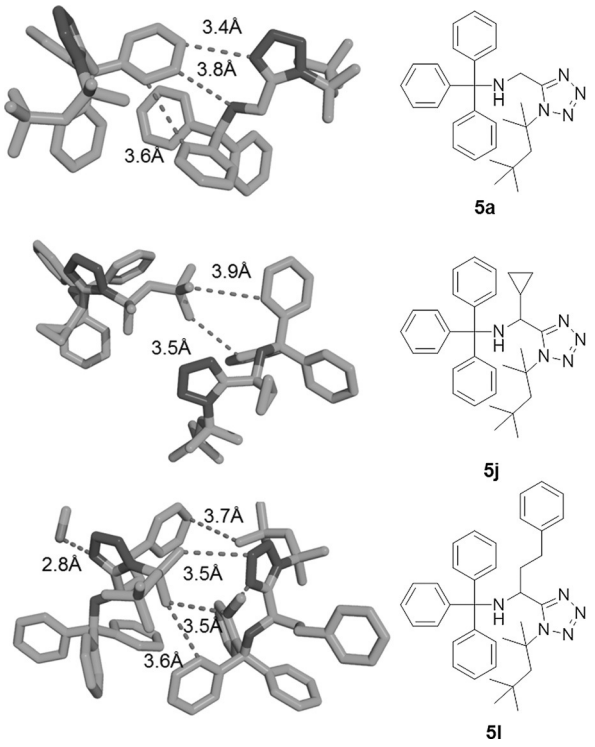


Figure 2. Molecular structures of compounds **5a**, **5j**, and **5j** in the solid state illustrating the crowdedness of the trityl-protected products and showing some short intermolecular atom–atom contacts. The crystal contacts are mostly governed by hydrophobic interactions. In **5l**, a co-crystallized methanol molecule forms a hydrogen bond to N4 of the tetrazole moiety. Thermal ellipsoids are drawn at the 50% probability level.

116.3, 53.0, 47.2, 43.3, 31.7, 25.9, 18.9 ppm; MS (ESI) *m/z* calculated $[M+H]^+$: 193.12; found $[M+H]^+$: 193.3.

Deprotecting procedure A using 1.25 M HCl in ethanol

Trityl group protected α-amino tetrazoles (**5a–l** and **5w**; 0.5 mmol) was dissolved in ethanol containing 1.25 M HCl (7.6 mL) in a round-bottomed flask with a magnetic stir bar. The reaction mixture was heated at reflux overnight. The solvent was evaporated under reduced pressure and the crude product was purified by column chromatography on silica eluting with CH_2Cl_2 (**6a–c**, **6e–l**) and methanol or on a Dowex 50 cation-exchange column eluted with 2N aqueous NH_3 (**6d**, **6w**).

(1*H*-Tetrazol-5-yl)methanamine hydrogen chloride (6a): The product was obtained as a light yellow oil (35 mg, 51%). $R_f = 0.41$ (CH_2Cl_2:MeOH 1:1); ^{1}H NMR (500 MHz, [D$_4$]methanol): $\delta = 4.40$ ppm (d, $J = 3.4$, 2H); ^{13}C NMR (126 MHz, [D$_4$]methanol): $\delta = 155.9$, 34.7 ppm; MS (ESI) *m/z* calculated $[M+H]^+$: 100.06; found $[M+H]^+$: 100.1.

Deprotecting procedure B using 10% Pd on charcoal

Benzyl group protected α-amino tetrazoles (**5n**, **5o′**, **5p–s**; 0.5 mmol) was dissolved in methanol (2 mL) containing 10% Pd on charcoal (500 mg) in a round-bottomed flask with a magnetic stir bar. The reaction mixture was stirred overnight at room temperature. The solid was filtered and the filtrate was concentrated under reduced pressure. The residue was purified on a Dowex 50 cation-exchange column eluted with 2N aqueous NH_3.

3-Amino-3-(1*H*-tetrazol-5-yl)propanoic acid (6n): The product was obtained as a light yellow solid (20 mg, 26%). $R_f = 0.89$ (CH_2Cl_2: MeOH 1:1); ^{1}H NMR (300 MHz, D_2O): $\delta = 4.98$ (dd, $J = 8.6$, 5.3 Hz, 1H), 3.04–2.88 ppm (m, 2H); ^{13}C NMR (75 MHz, D_2O): $\delta =$ 176.3, 44.8, 38.0, 25.8 ppm; MS (ESI) *m/z* calculated $[M+H]^+$: 171.09; found $[M+H]^+$: 171.2.

Deprotecting procedure using multiple deprotection

4-Amino-4-(1*H*-tetrazol-5-yl)butanamide (6m): 4-(1-Benzyl-1*H*-tetrazol-5-yl)-4-(tritylamino)butanamide (**5m′**; 70 mg, 0.14 mmol) was dissolved in methanol (0.5 mL). Then TFA (20 μL, 0.28 mmol) was added. The reaction mixture stirred for 30 min at room temperature. When the reaction was completed, 10% Pd on charcoal (139 mg) was added to the reaction mixture, which was then stirred overnight at room temperature under H_2. The reaction mixture was then concentrated under reduced pressure and the residue was purified by using a Dowex 50 cation-exchange column eluted with 2N aqueous NH_3. The product was white solid (14 mg, 60%). $R_f = 0.5$ (CH_2Cl_2:MeOH 1:1); ^{1}H NMR (300 MHz, D_2O): $\delta = 5.21$–5.11 (m, 0.5H), 4.93–4.83 (m, 0.5H), 2.76–2.50 (m, 1H), 2.42–2.20 (m, 2H), 2.15–1.95 ppm (m, 1H); ^{13}C NMR (126 MHz, D_2O): $\delta =$ 181.5, 175.6, 158.2, 155.9, 48.6, 29.0, 26.6 ppm; MS (ESI) *m/z* calculated $[M+H]^+$: 171.09; found $[M+H]^+$: 171.2.

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