Progress Towards the Total Synthesis of Yaku'amide A

Zhiwei Ma
Brigham Young University

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Progress Towards the Total Synthesis of Yaku’amide A

Zhiwei Ma

A dissertation submitted to the faculty of
Brigham Young University
in partial fulfillment of the requirements for the degree of

Doctor of Philosophy

Steven L. Castle, Chair
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July 2015

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ABSTRACT

Progress Towards the Total Synthesis of Yaku’amide A

Zhiwei Ma
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Doctor of Philosophy

The synthetic progress towards yaku’amide A is described. The study leads to development of new synthetic methodologies. Base-free regioselective aminohydroxylation is convenient to deliver β-\textit{tert}-hydroxyamino acids. A sequence consisting of alkylative esterification, Martin sulfurane mediated \textit{anti} dehydratation, a tandem azide reduction–O→N acyl transfer allows the rapid access of \textit{E}- and \textit{Z}-dehydroisoleucine-containing peptides from β-\textit{tert}-hydroxyisoleucine derivatives. Those methods are effective in constructing complicated peptides and advanced subunits of yaku’amide A.

Keywords: yaku’amide A, base-free regioselective aminohydroxylation, β-\textit{tert}-hydroxy amino acids, dehydroamino acids, dehydroisoleucine, Martin sulfurane, \textit{anti} dehyration, azide reduction, O→N acyl transfer.
First and foremost, I would like to thank my advisor Professor Steven L. Castle for his support, enthusiasm and patience in my development as an organic chemist. His insightful synthetic route of yaku’amide A has been great, highlighting the beauty of organic chemistry and motivating me to pursue higher goals in synthesis. His expertise in organic synthesis and dedication to the research are inspiring and teach me to work efficiently and diligently. His life philosophy has been a great influence on me as well.

I would like to thank Professors Merritt B. Andrus, Matt A. Peterson and David J. Michaelis for their kindness and expertise in discussions on dozens of tricky synthesis problems. Professor Paul B. Savage’s philosophy of “becoming the one who knows their project the most in the world” has been encouraging me all the way through my graduate study. Professors Joshua L. Price and Roger G. Harrison have been great source of motivation.

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being together to fill the lab life with diligent work and liveliness. All of the lab-mates deserve my gratitude for their help in improving my language.

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<td>Bn</td>
<td>Benzyl</td>
</tr>
<tr>
<td>Boc</td>
<td>tert-Butyloxy carbonyl</td>
</tr>
<tr>
<td>t-Bu or tBu</td>
<td>tert-Butyl</td>
</tr>
<tr>
<td>Burgess Reagent</td>
<td>Methyl N-(triethylammoniumsulfonyl)carbamate</td>
</tr>
<tr>
<td>Cbz</td>
<td>Carbobenzyloxy</td>
</tr>
<tr>
<td>CH$_2$Cl$_2$</td>
<td>Dichloromethane</td>
</tr>
<tr>
<td>COMU</td>
<td>(1-Cyano-2-ethoxy-2-oxoethylidenamino)oxydimethylamino-morpholino-carbenium hexafluorophosphate</td>
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<td>18-crown-6</td>
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<td>DABCO</td>
<td>1,4-Diazabicyclo[2.2.2]octane</td>
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<td>DBN</td>
<td>1,5-Diazabicyclo[4.3.0]non-5-ene</td>
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<tr>
<td>DBU</td>
<td>1,8-Diazabicyclo[5.4.0]undec-7-ene</td>
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<td>DCC</td>
<td>Dicyclohexyl Carboxiimide</td>
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<td>DEPBT</td>
<td>3-(Diethoxyphosphoryloxy)-1,2,3-benzotriazin-4(3H)-one</td>
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<tr>
<td>DIBAL</td>
<td>Diisobutylaluminum Hydride</td>
</tr>
<tr>
<td>DMF</td>
<td>Dimethylformamide</td>
</tr>
<tr>
<td>DMP</td>
<td>Dess-Martin Periodinane</td>
</tr>
<tr>
<td>DMSO</td>
<td>Dimethyl Sulfoxide</td>
</tr>
<tr>
<td>dr</td>
<td>diastereomeric ratio</td>
</tr>
<tr>
<td>ee</td>
<td>Enantiomeric Excess</td>
</tr>
<tr>
<td>EDC or EDCI</td>
<td>1-Ethyl-3-(3-dimethylamino propyl)carbodiimide</td>
</tr>
<tr>
<td>EDCI•HCl</td>
<td>1-Ethyl-3-(3-dimethylamino propyl)carbodiimide Hydrochloride</td>
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<tr>
<td>HOAt</td>
<td>7-Aza-1-hydroxybenzotriazole</td>
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<td>HOBt</td>
<td>1-Hydroxybenzotriazole</td>
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<td>Hünig's Base</td>
<td>Diisopropylethylamine</td>
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<td>HMBC</td>
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<td>LAH</td>
<td>Lithium Aluminum Hydride (LiAlH$_4$)</td>
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<td>Lindlar Catalyst</td>
<td>Pd on CaCO$_3$/PbO</td>
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<td>MS</td>
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<tr>
<td>Ms</td>
<td>Methanesulfonyl (Mesyl, CH$_3$SO$_2$)</td>
</tr>
<tr>
<td>NMO</td>
<td>N-Methylmorpholine-N-oxide</td>
</tr>
<tr>
<td>NMR</td>
<td>Nuclear Magnetic Resonance</td>
</tr>
<tr>
<td>PhthN</td>
<td>Phthalimido</td>
</tr>
<tr>
<td>Ph</td>
<td>Phenyl</td>
</tr>
<tr>
<td>PyBOP</td>
<td>Benzo triazol-1-yl oxy-tri pyrrolidinophosphonium hexafluorophosphate</td>
</tr>
<tr>
<td>Rf</td>
<td>Retention Factor (chromatography)</td>
</tr>
<tr>
<td>TBAF</td>
<td>Tetra-$n$-buty lammonium fluoride</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Full Form</td>
</tr>
<tr>
<td>--------------</td>
<td>-----------</td>
</tr>
<tr>
<td>TBDPS</td>
<td>tert-Butyldiphenylsilyl</td>
</tr>
<tr>
<td>TBS (TBDMS)</td>
<td>tert-Butyldimethylsilyl</td>
</tr>
<tr>
<td>TES</td>
<td>Triethyldimethylsilyl</td>
</tr>
<tr>
<td>Tf</td>
<td>Triflate (CF$_3$SO$_2$)</td>
</tr>
<tr>
<td>TFA</td>
<td>Trifluoroacetic acid</td>
</tr>
<tr>
<td>THF</td>
<td>Tetrahydrofuran</td>
</tr>
<tr>
<td>TLC</td>
<td>Thin Layer Chromatography</td>
</tr>
<tr>
<td>TMG</td>
<td>1,1,3,3-Tetramethylguanidine</td>
</tr>
<tr>
<td>TPAP</td>
<td>Tetra-$n$-propylammonium Perruthenate</td>
</tr>
<tr>
<td>UV</td>
<td>Ultraviolet spectroscopy</td>
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* Stereocenters marked by asterisks possess the indicated relative, not absolute stereochemistry.
Chapter 1. **YAKU’AMIDE A**

1.1 **Isolation and biological activity**

The acyclic peptides yaku’amide A and B were isolated from the *Ceratopsision* sponge collected at a depth of 150 m from the East China Sea, by Matsunaga et al. in 2010. These linear tetradecapeptides feature unusual molecular architecture such as rare N-terminal acyl and C-terminal amino groups, and they are rich in α,β-dehydroamino acid (ΔAA) and β-hydroxyamino acid (β-OHAA) residues, including Z-dehydroisoleucine (ΔIle), E-ΔIle, and dehydrovaline (ΔVal, Figure 1.1). It is noteworthy that the Z-ΔIle structure was unprecedented in natural products. The initially proposed structures of the peptides were based on extensive nuclear magnetic resonance spectroscopic analysis (\(^{1}\)H, \(^{13}\)C, COSY, HMBC, HMQC) and chemical degradations. The C-4 configuration of the N-terminal acyl subunit was undefined. In 2013, Inoue et al. synthesized the proposed structure of yaku’amide A and determined the C-4 S-configuration. In June 2015, a new structure of the yaku’amides was determined by Inoue et al., which showed that the originally proposed sequence of the neighboring L- and D-β-OHVal and L- and D-Val residues were switched. Possibly, the chemical degradation products were not characterized correctly during the original structure elucidation step. To simplify the naming, the originally determined structures will be represented as yaku’amide A' (1, Figure 1.1) and B' (2) and the corrected structures as yaku’amide A (3) and B (4) in this thesis.
Biological tests showed the potent anticancer activity of the yaku’amides. The IC₅₀ values for yaku’amide A and B against P388 murine leukemia cells are 14 and 4 ng/mL, respectively. The inhibition profile of yaku’amide A towards a panel of 39 human cancer cell lines (JFCR39) is unique compared to 38 known anticancer drugs that have been tested in this panel. Thus, it is believed that yaku’amide A possesses a novel mode of action to kill cancer cells.

However, the isolation of this natural product in minute quantities (ca. 1 mg) hinders further detailed study to elucidate its anticancer mechanism. Therefore, it is vital to synthesize yaku’amide A in the lab for further anticancer studies.

Figure 1.1 Yaku’amide A’ and B’; yaku’amide A and B.

1.2 Prior art for the synthesis of Z- and E-ΔIle

α,β-Dehydroisoleucine (ΔIle) is a key component of several bioactive peptide natural products. The E-ΔIle-containing phomopsins have been the focus of several research groups, and significant effort has been devoted to achieving a stereoselective construction of E-ΔIle. In their efforts toward the total synthesis of phomopsins, the Wandless and Joullié groups independently developed stereospecific dehydrations of β-hydroxyisoleucine (β-OHIle) derivatives for the construction of E-ΔIle. Wandless et al. devised a two-step protocol involving
the DBU-promoted anti elimination of a cyclic sulfamidite intermediate 6 obtained from treatment of β-OHlIle-containing dipeptide 5 with SOCl₂. Joullié et al. developed a Cu(OTf)₂-catalyzed and EDC-promoted syn elimination of β-OHlIle derivative 9 (Scheme 1.1). Dipeptide 8 is a key component for the natural products phomopsin A and B. During the course of converting the ΔIle-containing esters 7 and 10 to the carboxylic acid 8, the backbone amide protection was performed by both groups to facilitate the peptide chain elongation without alkene isomerization. This extra protection step is mandatory due to the facile formation of an azlactone intermediate 12, which is highly enolizable and leads to the alkene isomerized coupling product 14 (Scheme 1.1).³

Scheme 1.1 Prior art for ΔIle synthesis (Wandless and Joullié) and the azlactone formation.

In the course of synthesizing yaku'amide A', Inoue and co-workers innovatively utilized Buchwald’s Cu-catalyzed cross-coupling to deliver the key dehydroamino acids.⁷ The original reaction conditions were optimized to fuse alkenyl iodides and primary amides to construct Z-
and E-ΔIle as well as ΔVal. For instance, primary amide 15 was coupled to vinyl iodide 16 under the DMEDA–Cs₂CO₃–30 mol % CuI conditions, exclusively delivering E-ΔIle containing dipeptide 17 (Scheme 1.2). However, possibly due to functional group incompatibilities, protected primary alcohols were used in the cross-coupling instead of the more convenient amides or esters. Upon TBDPS removal from 17, the primary alcohol was released and further oxidized to carboxylic acid 18 via a SO₃·Py–Pinnick oxidation sequence. Based on the work of Wandless and Joullié, it is not surprising that the backbone amide of 18 was protected to effect an isomerization-free peptide coupling. Similarly, dipeptides 20 and 21 as well as pentapeptide 24 were delivered via this powerful transformation and used to synthesize yaku’amide A’. It is noteworthy that C-terminal pentapeptide 24 was constructed via the coupling of enamide 22 and peptide-like iodide 23.

Scheme 1.2 Prior art for ΔIle synthesis (Inoue).

1.3 Prior art for the synthesis of β-OHIle and β-OHVal

Besides dehydroamino acids, β-hydroxyamino acids are the other key components of yaku’amide A. Previously, asymmetric syntheses of β-OHVal and β-OHIle have been achieved
using chiral pool starting materials, which were employed in Inoue’s synthesis of yaku’amide A' (vide infra).

Lubell et al. used a Grignard addition of methylmagnesium bromide to the commercially available \( N\)-(tert-Butoxycarbonyl)-D-serine methyl ester 25 followed by a one-step oxidation to convert the primary alcohol to the carboxylic acid L-\( \beta \)-OHVal 27 (Scheme 1.3).\(^8\) The similar L-serine derivative can be used to deliver D-\( \beta \)-OHVal. Therefore, an enantioselective synthesis of both \( \beta \)-OHVal isomers can be achieved in 4 steps from the protected serine derivatives or in 8 steps from D- and L-serine.

\[
\begin{align*}
&\text{BocHN} & \text{CO}_2\text{Me} & \rightarrow & \text{BocHN} & \text{OH} \\
&25 & \text{N-Boc-D-serine methyl ester} & \rightarrow & 26 & \text{L-\( \beta \)-OHVal}
\end{align*}
\]

**Scheme 1.3** Prior art for L-\( \beta \)-OHVal synthesis.

Guanti et al. developed a 7-step protocol to access (2S,3R)-\( \beta \)-OHlle from D-Serine 28.\(^9\) The Grignard addition of ketone 29 constructed the stereocenter of the tertiary alcohol 30 with high diastereoselectivity due to chelation control (Scheme 1.4).

\[
\begin{align*}
&\text{HO} & \text{OH} & \text{NH}_2 \\
&28 & \text{D-Serine} & \rightarrow & \text{BocN} & \text{EtMgBr} \\
& & & \rightarrow & \text{HO} & \text{OH} \\
& & & \text{BocN} & \text{O} & \rightarrow & \text{BocHN} & \text{O}
\end{align*}
\]

**Scheme 1.4** Prior art for (2S,3R)-\( \beta \)-OHlle synthesis.

### 1.4 Inoue’s synthesis of yaku’amide A’

To furnish yaku’amide A', Inoue et al. also developed a method to access both enantiomers of the \( N \)-terminal acyl group (NTA). To simplify the description, only the
construction of NTA with correct C4-S stereochemistry will be discussed here. Evans asymmetric aldol reaction was utilized to selectively deliver the crucial C4 stereochemistry. Aldehyde 33 was obtained from diol 32 via monobenzylation and oxidation, which was reacted with the boron enolate of (R)-(−)-4-Benzyl-3-propionyl-2-oxazolidinone to deliver the syn-aldol adduct 34 (Scheme 1.5). Reductive cleavage of the chiral auxiliary successfully converted 34 into a diol. Via the combination of protecting group manipulation and carbon-chain elongation, diol 37 was obtained and further oxidized to the N-terminal carboxylic acid 38. It is noteworthy that the mild AZADO−PhI(OAc)_2 conditions successfully oxidized 37 to β-ketone carboxylic acid 38 in one pot, without concomitant C4-epimerization or C1-decarboxylation.

![Scheme 1.5 Inoue’s synthetic route to the N-terminal acyl group.](image)

The successful syntheses of all the required fragments of yaku’amide A set the stage to furnish the natural product. Inoue et al. utilized a “right-to-left” strategy to assemble those fragments due to the availability of the prepared starting materials (all fragments were ending with free carboxylic acids), which also lead to a relatively linear synthesis rather than a highly convergent synthesis. To start the peptide chain elongation process, the Boc group of the C-terminal pentapeptide 24 was cleaved and the resulting amine was coupled to dipeptide 21 to deliver the heptapeptide 39 (Scheme 1.6). Six sets of similar operations were able to install all the other fragments and deliver yaku’amide A'. The C-4 epimer of yaku’amide A' was prepared
in the same fashion. Spectral comparison of synthetic isomers to natural isolated yaku’amide A determined the C-4 S stereochemistry and verified the success of their total synthesis. It is interesting that the $^1$H and $^{13}$C NMR spectra of yaku’amide A' and yaku’amide A are apparently very similar, although they differ in configuration at four stereocenters. Two different coupling systems were used extensively in this project, namely PyBop–HOAt–$i$-Pr$_2$NEt and COMU–2,4,6-collidine, to eliminate the possible epimerization.

![Chemical structure](image)

**Scheme 1.6** Inoue’s route to the total synthesis of yaku’amide A'.

### 1.5 Conclusion

Inoue’s work features a highly efficient Cu(I)-catalyzed cross-coupling reaction and a novel route to synthesize the N-terminal acyl group. However, the efficiency of this pioneering work suffers from lengthy synthesis of β-OHAAs, inefficient functional group manipulations in the ΔAAs construction, and backbone amide protection. Thus, this work totals 86 steps with a 25-step longest linear sequence. There is a clear need for a more efficient synthesis of yaku’amide A.
To obtain this scarce but promising anticancer agent in a more practical fashion, development of a concise synthesis of Z- and E-\(\Delta\text{Ile}\) and \(\beta\)-OHAAs would be crucial and urgent. Therefore, we initiated research efforts in pursuit of an efficient and backbone amide protection-free stereoselective \(\Delta\text{Ile}\) synthesis.

1.6 References


(3) (a) Personal communication between Professor Steven L. Castle and Professor Masayuki Inoue (June 2015); (b) Kuranaga, T.; Mutoh, H.; Sesoko, Y.; Goto, T.; Matsunaga, S.; Inoue, M. J. Am. Chem. Soc. 2015, http://pubs.acs.org/doi/abs/10.1021/jacs.5b05550

(4) For a review of natural products containing \(\Delta\text{Ile}\) and other bulky dehydroamino acids, see: Jiang, J.; Ma, Z.; Castle, S. L. Tetrahedron 2015, 71, 5431–5451.


Chapter 2. **REGIOSELECTIVE AMINOHYDROXYLATION**

2.1 **Introduction to aminohydroxylation methods**

The unusual amino acid residues are the most challenging substructures incorporated in yaku’amide A, and the synthesis of those AAs dictates the efficiency of the total synthesis of the natural product. Wandless and Joullie’s efforts in the ΔIle synthesis enlightened us to construct ΔAA residues via a similar dehydration of β-tert-OH amino acids (chapter 1). Among reported methods to synthesize β-tert-OH amino acids, the alkene oxyamination chemistry was chosen due to its straightforwardness.\(^1\) The Sharpless aminohydroxylation is well-known for its wide application in asymmetric synthesis of β-OH 1,2-amino alcohols.\(^2\) However, this protocol is not applicable to our project due to the scope limitation of mono- and dissubstitued alkenes and the problem of regioselectivity.\(^3\)

To overcome the common regioselectivity problem existing in the intermolecular oxyamination reactions, Donohoe et al. devised a tethered aminohydroxylation method to access β-OH amino acids with fully controlled regioselectivity via an intramolecular pathway.\(^4\) This strategy was attempted in our lab on a trisubstituted allylic alcohol and delivered the desired product 48 (Scheme 2.1). However, multi-step operations were required to tether the nitrogen source to alkene 44 and then remove the tether after the aminohydroxylation. This problem combined with the low overall yield and the difficulty in the aqueous extraction of the polar amino diol 48 made us abandon this route.
Recently, Luxenburger and co-workers developed a base-free aminohydroxylation method to access the β-OHAA(s) from mono- or disubstituted alkenes. This method features an intermolecular regioselective syn addition of respective nitrogen source and hydroxyl group to the alkenes using a benzoyloxycarbamate. We envisioned the possibility of broadening the scope to the more challenging trisubstituted alkenes to deliver the β-tert-OHAA(s) that are present in yaku’amide A. We hypothesized that the bulkiness of the trisubstituted alkenes would act like a double-edged sword, bringing more steric hindrance but affording better regioselectivity to the aminohydroxylation reaction. The possible drawback could be circumvented via employing high catalyst loadings or higher reaction temperatures.

2.2 **Optimization of the aminohydroxylation conditions**

The study to verify our hypothesis was initiated with the aminohydroxylation of prenol 49. The reaction was fully regioselective under Luxenburger’s conditions (Table 2.1, entry 1). Thus, we further explored the reaction conditions using the model substrates prenol 49 and isoprenol 50 to obtain optimum conditions. The trisubstituted alkene prenol was chosen since its aminohydroxylation product 52 can be used as a ΔVal precursor. The details of our investigation are shown in Table 2.1. There was no obvious reactivity difference between the CH$_3$CN–H$_2$O or...
t-BuOH–H₂O solvent systems, so the former was chosen as a homogenous solvent system (entries 1 and 3). However, the N-Boc carbamate 51b showed slightly better reactivity than N-Cbz (75% versus 63%, entries 2 and 3). Due to the isolation convenience of UV-active compounds, we chose the N-Cbz carbamate 51a to perform our study. For the isoprenol 50, initial yield obtained was only 30%, possibly due to the extreme bulkiness of one alkene terminus. Analysis of the crude reaction mixture revealed that the hydrolysis of the carbamate 51a happened predominantly, delivering Cbz-NH₂ as the side product. Similar observations were made by Donohoe with tethered aminohydroxylations.⁴d To circumvent this problem, the use of base additives as ligands was explored and we were pleased to find that pyridine and DABCO both gave improved yields (entries 5–6). However, the use of other bases (i.e., NEt₃, DMAP, imidazole) did not induce any improvement, showing the significance of subtle changes in the ligand-promoted reactions.⁷ Finally, we were pleased to find that simple heating brought significant improvement and delivered 53 in 85% yield (entry 7). Thus, the conditions of entry 7 were chosen as the standard to investigate the scope of the aminohydroxylation.
Table 2.1 Optimization of aminohydroxylation conditions.

![Chemical structure](image)

<table>
<thead>
<tr>
<th>entry</th>
<th>alkene</th>
<th>R</th>
<th>ligand</th>
<th>product</th>
<th>yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>49</td>
<td>Cbz (51a)</td>
<td>none</td>
<td>52a</td>
<td>63%</td>
</tr>
<tr>
<td>2</td>
<td>49</td>
<td>Cbz</td>
<td>none</td>
<td>52a</td>
<td>63%</td>
</tr>
<tr>
<td>3</td>
<td>49</td>
<td>Boc (51b)</td>
<td>none</td>
<td>52b</td>
<td>75%</td>
</tr>
<tr>
<td>4</td>
<td>50</td>
<td>Cbz</td>
<td>none</td>
<td>53</td>
<td>30%</td>
</tr>
<tr>
<td>5</td>
<td>50</td>
<td>Cbz</td>
<td>pyr</td>
<td>53</td>
<td>61%</td>
</tr>
<tr>
<td>6</td>
<td>50</td>
<td>Cbz</td>
<td>DABCO</td>
<td>53</td>
<td>59%</td>
</tr>
<tr>
<td>7&lt;sup&gt;b&lt;/sup&gt;</td>
<td>50</td>
<td>Cbz</td>
<td>none</td>
<td>53</td>
<td>85%</td>
</tr>
</tbody>
</table>

<sup>a</sup>r-BuOH–H₂O 6:1 was used as a solvent.  
<sup>b</sup>Reaction was performed at 35 °C.

2.3 Investigation of the aminohydroxylation scope

With the optimal aminohydroxylation conditions in hand, we evaluated the reaction with several di- or trisubstituted alkenes, including allylic or homoallylic alochols and an enoate (Table 2.2). Single regioisomers were obtained in good yields from aminohydroxylations of allylic ether 54, enoate 56, and homoallylic alcohol 58 (Table 2.2, entries 1–3). Aminohydroxylations of bulkier trisubstituted alkenes 60, 62, and 64 effected the single regioisomers with moderate yields (entries 4–6). This selectivity could be ascribed to the steric differences between the two alkene carbons. Consistent with the study on isoprenol 50, conversion of methallyl alcohol 66 to the amino diol 67 happened smoothly at rt in good yield (entry 7). Aminohydroxylation of the monosubstituted alkene 68 predominantly delivered the amino diol 69 at 70% yield with 10% cogeneration of the meso isomer (entry 8). A 10 mol % loading of OsO₄ greatly improved the yields with several highly bulky substrates (entries 2, 5, and 6). The respective products 57, 63, and 65 can be used as precursors to ΔVal or ΔIle.
Table 2.2 Reaction scope of regioselective aminohydroxylation.

\[
\begin{array}{cccc}
\text{entry} & \text{alkene} & \text{product [temp., yield]} & \text{entry} & \text{alkene} & \text{product [temp., yield]} \\
1 & \text{CO}_2\text{Et} & \text{OH} & 5 & \text{OH} & \text{62} \\
2 & \text{OH} & \text{CBzHN} & 6 & \text{OH} & \text{64} \\
3 & \text{Ph} & \text{OH} & 7 & \text{OH} & \text{66} \\
4 & \text{OH} & \text{CBzHN} & 8 & \text{OH} & \text{68} \\
5 & \text{OMe} & \text{OH} & 63 & \text{OH} & \text{65} \\
6 & \text{OMe} & \text{OH} & 64 & \text{OH} & \text{67} \\
7 & \text{OMe} & \text{OH} & 65 & \text{OH} & \text{67} \\
8 & \text{OMe} & \text{OH} & 66 & \text{OH} & \text{68} \\
\end{array}
\]

\textsuperscript{a} 10 mol \% OsO\textsubscript{4} was used. \textsuperscript{b} 10 mol \% loading of OsO\textsubscript{4} was used at 35 °C.
\textsuperscript{c} A 6.7:1 mixture of regioisomers was obtained.

2.4 ΔVal-containing peptide synthesis

With the success in the synthesis of β-OHAAs, we initiated the model study of ΔVal-containing peptide synthesis. β-OHVal 57 was converted to the respective amine via Pd/C catalyzed hydrogenolysis, and the amine was coupled to Cbz-glycine, delivering dipeptide 70 with overall yield of 80\% (Scheme 2.2). Due to the robustness of the Cbz group, high pressures of hydrogen (450 PSI) were required to maintain a fast cleavage rate. Convenient accessible dipeptide 70 was chosen as the model compound to investigate the dehydration reactions.
To dehydrate the tertiary alcohol 70 regioselectively, we screened many conditions reported in the literature (more than 100 different variants), and the representative conditions and results are shown in Table 3. The beginning effort to convert the tertiary alcohol to a mesylate or tosylate was unsuccessful due to facile elimination in situ, delivering both the desired product 71a and its regioisomer 71b (entries 1–2, Table 2.3). Attempted one-pot elimination without separation of the intermediates gave similar results. Acetic anhydride related conditions delivered mono- or di-acylated products instead of any further elimination product, and these acetates could not be eliminated under TBAF or DBU conditions (entries 3–5). The Burgess dehydrating reagent gave both regioisomers as well, and attempts to decrease the temperature to improve the regioselectivity were unsuccessful (entry 6). Joullié’s EDCI-promoted Copper(II) Lewis acid-catalyzed dehydration was appealing due to its high yielding and one-step operation. However, the application of this method to our substrate was unsuccessful (entries 7–8). Significant effort was devoted to optimizing this method via screening different combinations of solvents, Lewis acids and temperatures, but the best yield we obtained was 30%. Also, this yield was difficult to reproduce (entries 9–10). The Mitsunobu-type conditions and Lewis acid-catalyzed dehydration conditions caused decomposition or returned the starting material (entries 11–14). Dehydrating reagents SOCl₂, SO₂Cl₂ and POCl₃ were also not able to deliver the desired product 71a (entries 15–17). Interestingly, the Swern oxidation conditions delivered the elimination product at 21% yield (entry 18). Wandless conditions delivered the desired ΔVal-
containing peptide 71a at 47% yield (entry 19). Ultimately, we found Martin sulfurane (Table 2.3, shown in red) exhibited superior activity and delivered the dipeptide 71a in 80% yield (entry 20). This success demonstrates the feasibility of our proposed oxyamination-derivatization-dehydration sequence in producing the crucial ΔAA-containing peptides.

Table 2.3 Investigation of dehydration conditions.

<table>
<thead>
<tr>
<th>entry</th>
<th>reaction condition</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>MsCl, NEt₃; DMAP or DBU or Pyr</td>
<td>71a and 71b</td>
</tr>
<tr>
<td>2</td>
<td>Tf₂O, Pyr, CH₂Cl₂ -78 °C or 0 °C</td>
<td>71a and 71b</td>
</tr>
<tr>
<td>3</td>
<td>Ac₂O, DMAP, NEt₃</td>
<td>N- and O-acylation product</td>
</tr>
<tr>
<td>4</td>
<td>Ac₂O, 100 °C</td>
<td>O-acylation product</td>
</tr>
<tr>
<td>5</td>
<td>Sc(OTf)₃, Ac₂O, 60 °C</td>
<td>N- and O-acylation product</td>
</tr>
<tr>
<td>6</td>
<td>Burgess's Reagent, -30 °C or 0 °C or rt</td>
<td>71b</td>
</tr>
<tr>
<td>7</td>
<td>EDC, CuCl₂, Toluene, 80 °C</td>
<td>Decomposition</td>
</tr>
<tr>
<td>8</td>
<td>EDC, Cu(OTf)₂, THF/ DMF, 60 °C</td>
<td>71a 33% (Not reproducible)</td>
</tr>
<tr>
<td>9</td>
<td>Yb(OTf)₃, EDC, THF, 70 °C</td>
<td>71a 30% (Not reproducible)</td>
</tr>
<tr>
<td>10</td>
<td>Sc(OTf)₃ or La(OTf)₃ or Y(OTf)₃, EDC, THF, 70 °C</td>
<td>Decomposition</td>
</tr>
<tr>
<td>11</td>
<td>I₂, PPh₃, CH₂Cl₂, rt or 60 °C</td>
<td>Decomposition</td>
</tr>
<tr>
<td>12</td>
<td>BF₃OEt₂, CH₂Cl₂, rt</td>
<td>No reaction</td>
</tr>
<tr>
<td>13</td>
<td>BF₃THF, CH₂Cl₂, rt</td>
<td>Decomposition</td>
</tr>
<tr>
<td>14</td>
<td>CeCl₃+H₂O, NaI, CH₃CN, rt or reflux</td>
<td>No reaction</td>
</tr>
<tr>
<td>15</td>
<td>POCl₃, Pyr or NEt₃, 0 °C or rt</td>
<td>Decomposition</td>
</tr>
<tr>
<td>16</td>
<td>SOCl₂, 2,6-lutidine, CH₂Cl₂</td>
<td>Sulfonamide intermediate</td>
</tr>
<tr>
<td>17</td>
<td>SO₂Cl₂, 2,6-lutidine, CH₂Cl₂</td>
<td>Decomposition</td>
</tr>
<tr>
<td>18</td>
<td>DMSO, (COCl)₂, -78 °C; TBAF</td>
<td>71a 21% (brsm: 32%)</td>
</tr>
<tr>
<td>19</td>
<td>SOCl₂, CH₂Cl₂, NEt₃, -78°C; DBU</td>
<td>71a 47%</td>
</tr>
<tr>
<td>20</td>
<td>Martin sulfurane, 50 °C, CHCl₃</td>
<td>71a 80%</td>
</tr>
</tbody>
</table>

2.5 Conclusion

During the course of synthesizing dehydroamino acids, a base-free regioselective aminohydroxylation method was developed, which allows rapid access to β-tert-OHAAAs. The
application of this method was demonstrated via synthesizing a model ΔVal-containing dipeptide. The dehydration process was mediated by the Martin sulfurane dehydrating reagent. This progress set the stage for the investigation of a synthetic method to access ΔIle-containing peptides stereoselectively.

2.6 References


Chapter 3. THE CONSTRUCTION OF ΔAA-CONTAINING TRIPEPTIDES

In Chapter 2, we demonstrated that the ΔVal-containing peptide can be accessed via dehydration of a β-OHVal-containing precursor using Martin sulfurane. In the following stage, we pursued a concise incorporation of tetrasubstituted dehydroamino acids into peptides.

3.1 Attempt of direct dehydration of tripeptide

With the successful synthesis of the ΔVal-containing dipeptide, we considered the possibility of direct dehydration of the tripeptide 73 containing β-OHVal as the middle segment was synthesized from ester 69 via a saponification–coupling sequence (Scheme 3.1).

![Scheme 3.1 Synthesis of amide 73.](image)

The best dehydration conditions from Chapter 2 were tested on this more challenging substrate (Table 3.1). Disappointingly, EDCI-Lewis acid combinations did not deliver any detectable product, and only recovered starting material or decomposition was observed (Table 3.1, entries 1 and 2). The Wandless conditions were unsuccessful, as well as the POCl₃/Pyr conditions (entries 3 and 4). Various combinations of temperature and equivalents of the Martin sulfurane were screened, but all led to recovered starting material or decomposition (summarized...
Therefore, we abandoned the dehydration of β-hydroxy amino acids embedded in peptides and pursued other alternatives.

Table 3.1 Dehydration study of amide 73.

<table>
<thead>
<tr>
<th>entry</th>
<th>condition</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.2–0.9 eq Yb(OTf)$_3$, EDC, 60 °C</td>
<td>Starting material</td>
</tr>
<tr>
<td>2</td>
<td>0.2–2.0 eq Cu(OTf)$_2$, EDC, 60 °C</td>
<td>Starting material or decomposition</td>
</tr>
<tr>
<td>3</td>
<td>i. SOCl$_2$, NEt$_3$, –78 °C; ii. DBU</td>
<td>Decomposition</td>
</tr>
<tr>
<td>4</td>
<td>POCl$_3$, Pyr. 0 °C to rt</td>
<td>Decomposition</td>
</tr>
<tr>
<td>5</td>
<td>1.0–3.3 eq Martin sulfurane, rt–80 °C</td>
<td>No reaction or decomposition</td>
</tr>
</tbody>
</table>

3.2 Aminohydroxylation using a new benzyl mesyloxy carbamate

The failure in the direct dehydration of 73 as well as pioneering efforts of Wandless and Joullié showed the importance of obtaining a ΔIle ester intermediate en route to ΔIle-containing peptides. Therefore, we focused on the development of a stereospecific dehydration of β-OHIle substrates 77 and 80, which could be delivered via an aminohydroxylation–coupling sequence from alkenes 75 and 78 (Scheme 3.2). Similar to the aminohydroxylation of isoprenol in Chapter 2, Cbz-NH$_2$ was obtained predominantly when benzyl $p$-chlorobenzyloxycarbamate 51a (Chapter 2) was used to aminohydroxylate the bulky and electron-deficient alkenes 75 and 78. Herein, a new benzyl mesyloxy carbamate (CbzHN–OMs) was used that delivered aminohydroxylation adducts 76 and 79 with high efficiency and suppressed Cbz-NH$_2$ generation. The improved activity of CbzHN–OMs could be attributed to the decreased basicity of –OMs leaving group compared to the original 4-chlorobenzooyl group. A related phenomenon was observed by Donohoe and co-workers in their tethered aminohydroxylation work. While the
development of a stereoselective dehydration towards Z- and E-ΔIle was underway in our lab, McLeod group reported the advantageous reactivity of sulfonyloxycarbamates in base-free aminohydroxylations. With the success of constructing 76 and 79, the backbone elongation strategy used in the ΔVal synthesis worked without incident, affording the model dipeptides 77 and 80 in good yields.

Scheme 3.2 Synthesis of β-OHIle-containing dipeptides 77 and 80.

3.3 Investigation of the Wandless dehydration protocol

Since the Martin sulfurane dehydrating reagent was well known for reacting via an E1-like mechanism with tertiary alcohols, we were not expecting a stereoselective dehydration to occur with this reagent. During the dehydration study outlined in Chapter 2, the Wandless’ SOCl₂–DBU 2-step protocol was the most promising method for delivering the ΔIle-containing peptides. Therefore, we used this method to pursue a backbone amide protection-free ΔIle synthesis. The moderate yield and stereoselectivity obtained on dehydration of 77 (Scheme 3.3) were not ideal but demonstrated the possibility of using this method for accessing ΔIle-containing peptides.
Scheme 3.3 *Anti* dehydration of tertiary alcohol 77 via Wandless method.

3.4 Broadening the Wandless protocol

With the positive results using Wandless chemistry, we considered the possibility of producing ΔIle-containing peptides via transamidation of an activated ester intermediate.

3.4.1 The investigation of pentafluorophenyl (C₆F₅) ester

The pentafluorophenyl (C₆F₅) ester was chosen to test our proposal due to its application in amide bond construction.⁶ From carboxylic acid 72, pentafluorophenyl ester 83 was obtained via EDCI•HCl–pentafluorophenol conditions. It was further converted to the sulfamidite intermediate by treating with SOCl₂, and two routes were investigated. Route A was a dead end since direct elimination of the sulfamidite 84 produced the azlactone 86 exclusively. In Route B, sulfamid 84 was treated with the free amine of glycine ethyl ester, delivering the advanced sulfamidite 85 via transamidation. Unfortunately, the following elimination afforded the desired product 83 and tertiary alcohol 73 as the side product. After screening various bases (TMG, DBU, DBN), the best yield obtained was 24% and the side product generation could not be circumvented. This result is not efficient enough for use in the future total synthesis of yaku’amide A.
3.4.2 The investigation of phenyl ester

To address the problem of the pentafluorophenyl ester cleavage during the elimination step, we switched to the more robust phenyl ester. We began with a similar saponification–coupling strategy to access the phenyl ester 89 from ester 72 (Scheme 3.5). However, various coupling conditions all led to dehydration of the tertiary alcohol or azlactone formation. Therefore, starting from alkene 88, an aminohydroxylation–hydrogenolysis–coupling sequence delivered dipeptide 89 (Scheme 3.5). Subsequently, alcohol 89 was converted to sulfamidite 90, setting the stage to test the transamidation–elimination sequence as well as the sequence with the reverse order. Disappointingly, azlactone was detected in both routes as well as significant decomposition of the starting material. Therefore, the phenyl ester chemistry was abandoned. The aminohydroxylation and Cbz cleavage steps were low yielding and only one attempt of this chemistry was performed, therefore, no exact yields were obtained for the reactions shown in Scheme 3.5 (This rule also applies to most of the investigative chemistry in the dissertation).
3.5 Attempts to elongate the dipeptide via carboxylic acid coupling

With the failure to combine the Wandless elimination protocol with transamidation, we examined the possibility of fusing the carboxylic acid with an amine using nontraditional coupling conditions to avoid formation of the azlactone intermediate. Several newly reported coupling methods were investigated on our substrate. The ZrCl$_4$-catalyzed coupling reaction with no activated ester intermediate showed the possibility to effect isomerization-free coupling. Therefore, the carboxylic acid 93 was obtained via saponification of 66 and subjected to the coupling conditions. However, product was not detected and the azlactone formation was observed during heating (Table 3.2, entry 1).\(^7\) In 2012, Ashfeld and co-workers reported a peptide coupling method via traceless Staudinger ligation (entry 2).\(^8\) This method featured an O→N acyl transfer to deliver a key phosphite ylide intermediate, which could be hydrolyzed to deliver the amide. Unfortunately, attempts of this method were fruitless and the azlactone side product was formed again. Recently, Sheppard and co-workers developed a B(OCH$_2$CF$_3$)$_3$-catalyzed amination process.\(^9\) The attempts with B(OCH$_2$CF$_3$)$_3$ resulted in formation of some amide 94 as evidenced by mass spectrometry. However, desired product could not be isolated.
from SiO$_2$ purification. NMR analysis of the crude reaction mixture also disclosed significant decomposition (entry 3).

**Table 3.2** Non-traditional coupling study.

<table>
<thead>
<tr>
<th>entry</th>
<th>condition</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>25 mol % ZrCl$_4$, NH$_2$Bn, 70 °C, 22 h</td>
<td>95 with decomposition</td>
</tr>
<tr>
<td>2</td>
<td>BnN$_3$, CIP(OEt)$_2$, NEt$_3$, 1,4-dioxane, 80 °C</td>
<td>95 with decomposition</td>
</tr>
<tr>
<td>3</td>
<td>B(OCH$_2$CF$_3$)$_3$, NH$_2$Bn, CH$_3$CN, 90 °C</td>
<td>Decomposition</td>
</tr>
</tbody>
</table>

Furthermore, failure in applying the Wandless protocol to pentafluorophenyl ester 83 and phenyl ester 89 led us to conduct an alkylative esterification on the carboxylic acid 93 to form a 2,2,2-trifluoroethyl ester 96. This attempt was successful and ester 96 was subsequently subjected to the transamination condition. The amide 94 was evidenced by mass spectrometry only when extra equivalents (>100 equiv) of benzyl amine were loaded into the reaction. Therefore, this is not practical for the future total synthesis due to the high cost of enantiomerically pure β-amino alcohols. In addition, the heavy base loading is a concern to cause azlactone formation or perform reversible conjugate addition to the alkene, and either pathway could lead to isomerization of the alkene.

**Scheme 3.6** Transamidation of 2,2,2-trifluoroethyl ester.
During the meantime, the saponification of Z-ΔIle-containing 82 was tested and it turned out to be a slow process. Careful analysis of the crude reaction mixture via NMR and MS revealed that the hydrolysis process was accompanied by alkene isomerization (ca. 2:1 dr), azlactone formation, or both (Table 3.3). This observation was different from Joullie’s report on isomerization-free hydrolysis of a similar substrate, showing that large reactivity variations could be caused by subtle structural differences. Disappointingly, those side reactions were impossible to circumvent after thoroughly tuning of reaction conditions (Table 3.3). The best explanation might be the bulkiness of the ester allowing reversible enolization to happen via γ-deprotonation, which scrambled the ΔIle stereochemistry. Finally, this saponification–nontraditional coupling strategy was abandoned due to the inaccessibility of the crucial carboxylic acid intermediates.
Table 3.3 Investigation of the saponification of ester 82.

<table>
<thead>
<tr>
<th>entry</th>
<th>condition</th>
<th>result (97a : 97b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.1 eq LiOH, EtOH–H₂O, 60 °C, 3 h, then rt, 15 h</td>
<td>2:1</td>
</tr>
<tr>
<td>2</td>
<td>1.0 eq LiOH, THF–EtOH–H₂O, 60 °C, 24–120 h</td>
<td>1:1</td>
</tr>
<tr>
<td>3</td>
<td>1.0 eq LiOH, EtOH–H₂O, 70 °C, 6 h</td>
<td>2.5:1</td>
</tr>
<tr>
<td>4</td>
<td>1.0 eq LiOH, THF–EtOH–H₂O, 70 °C, 3 h</td>
<td>1:1</td>
</tr>
<tr>
<td>5</td>
<td>1.5 eq LiOH, EtOH–H₂O, 60 °C, 3 h</td>
<td>2:1</td>
</tr>
<tr>
<td>6</td>
<td>1.5 eq LiOH, THF–EtOH–H₂O, 60 °C, 60 °C, 3 h</td>
<td>1.8:1</td>
</tr>
<tr>
<td>7</td>
<td>2.0 eq LiOH, EtOH–H₂O, 50 °C, 2 h</td>
<td>1.8:1</td>
</tr>
<tr>
<td>8</td>
<td>2.0 eq LiOH, THF–EtOH–H₂O, 50 °C, 2 h</td>
<td>1.8:1</td>
</tr>
<tr>
<td>9</td>
<td>0.95–2.0 eq LiOH, THF–MeOH–H₂O, 50 °C, 2 h</td>
<td>2:1–1:1</td>
</tr>
<tr>
<td>10</td>
<td>3.3 eq LiOH, 10 eq H₂O, CH₃CN, rt, 72 h</td>
<td>No reaction</td>
</tr>
<tr>
<td>11</td>
<td>CaCl₂, NaOH, isopropanol–H₂O, rt, 24 h (40 °C, 3 h)</td>
<td>82 (98 with 82)</td>
</tr>
<tr>
<td>12</td>
<td>5 eq LiOH (1M in H₂O), MeOH, rt, 3 d</td>
<td>1:1 with 82</td>
</tr>
<tr>
<td>13</td>
<td>NaOH, EtOH or Dioxane, rt, 24 h or 70 °C, 1 h</td>
<td>98 with 82</td>
</tr>
<tr>
<td>14</td>
<td>4.5–7.0 eq Me₃SnOH, (CH₂Cl)₂, 80 °C, 15 h</td>
<td>98 with 82</td>
</tr>
<tr>
<td>15</td>
<td>LiOOH</td>
<td>82</td>
</tr>
<tr>
<td>16</td>
<td>Lil, pyr. 130 °C</td>
<td>Decomposition</td>
</tr>
</tbody>
</table>

*The ratio was determined by analysis of the NMR of crude reaction mixture.

3.6 Transamidation of non-activated esters

Due to the incapability of performing isomerization-free saponification, the direct transamidation method was considered and investigated. Similarly, the ΔVal-containing methyl ester 99 (Table 3.4) was synthesized via Martin sulfurane mediated dehydration, and subjected to two popular transamidation conditions. The trimethyl aluminum-catalyzed transamidation did not proceed at room temperature or decomposed upon being heated (entry 1). The direct transamidation only happened with large equivalents loading of benzyl amine (entry 2), which is similar to previously tested 2,2,2-trifluoroethyl ester 96 (Scheme 3.6). Therefore, this transamidation pathway via methyl ester was concluded as failure.
Table 3.4 Transamidation of methyl ester.

<table>
<thead>
<tr>
<th>entry</th>
<th>reaction condition</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>AlMe₃, NH₂Bn, C₆H₅Cl, rt (50–80 °C)</td>
<td>⁹⁹ (Decomposition)</td>
</tr>
<tr>
<td>2</td>
<td>MeOH, NH₂Bn, rt, 3 d</td>
<td>⁹⁴</td>
</tr>
</tbody>
</table>

3.7 The attempts of performing thioester coupling chemistry

Recently, Aimoto reported a thioester-mediated amide synthesis protocol, and Inoue et al. utilized this strategy in the total synthesis of polytheonamide B. Encouraged by the feasibility of this method in challenging coupling reactions as well as the utility of thioesters in amide bond formation, we attempted to synthesize a thioester intermediate to investigate this indirect amide coupling pathway. However, the direct coupling from carboxylic acid ⁷₂ to thioester ¹⁰₁ was fruitless (Scheme 3.7). The longer aminohydroxylation–hydrogenolysis–coupling sequence delivered very minor amounts of alcohol ¹⁰₁ from alkene ¹⁰⁰. The low conversion was possibly caused by the incompatibility of the thioester moiety with the aminohydroxylation and hydrogenolysis conditions. Furthermore, the failure of the DBU-promoted elimination of sulfamidite ¹⁰₂ made us abandon this route as well.
Due to the fact that the Wandless protocol could not deliver high enough yields and stereoselectivities on our substrates, finding an efficient dehydration method with high selectivity was crucial and urgent to the total synthesis. We pursued useful dehydration method in parallel with an isomerization-free coupling pathway. With all possible known dehydration conditions tested, we decided to try Martin sulfurane dehydrating reagent, which we did not expect to be promising due to its propensity to promote E1 dehydration.\(^5\) Surprisingly, we found that it delivered Z- and E-dehydroisoleucine-containing peptides 104 and 105 from the alcohols 77 and 80, respectively, in high yields (Scheme 3.8). Crude NMR analysis of those stereoconvergent dehydration reactions showed that a single detectable isomer 104 or 105 was delivered from a pair of enantiomers (to the limits of detection by \(^1\)H NMR), and both alkene isomers are complementary to each other. This one-step dehydration protocol is convenient and powerful, delivering the best yields and diastereomeric ratios among all the tested conditions. This success broadens the application of our aminohydroxylation method, delivering isomerically pure ΔIle-containing peptides in only four chemical transformations from enoates 75 and 77 (Scheme 3.2). Although the cost is relatively high ($50/g), Martin sulfurane can be conveniently synthesized.

Scheme 3.7 The attempts of synthesizing alkene thioester 103.

3.8 Martin sulfurane dehydration

Due to the fact that the Wandless protocol could not deliver high enough yields and stereoselectivities on our substrates, finding an efficient dehydration method with high selectivity was crucial and urgent to the total synthesis. We pursued useful dehydration method in parallel with an isomerization-free coupling pathway. With all possible known dehydration conditions tested, we decided to try Martin sulfurane dehydrating reagent, which we did not expect to be promising due to its propensity to promote E1 dehydration.\(^5\) Surprisingly, we found that it delivered Z- and E-dehydroisoleucine-containing peptides 104 and 105 from the alcohols 77 and 80, respectively, in high yields (Scheme 3.8). Crude NMR analysis of those stereoconvergent dehydration reactions showed that a single detectable isomer 104 or 105 was delivered from a pair of enantiomers (to the limits of detection by \(^1\)H NMR), and both alkene isomers are complementary to each other. This one-step dehydration protocol is convenient and powerful, delivering the best yields and diastereomeric ratios among all the tested conditions. This success broadens the application of our aminohydroxylation method, delivering isomerically pure ΔIle-containing peptides in only four chemical transformations from enoates 75 and 77 (Scheme 3.2). Although the cost is relatively high ($50/g), Martin sulfurane can be conveniently synthesized.
from inexpensive commercially available starting materials (<$3/g) following Martin’s one-step protocol. 

Scheme 3.8 Martin sulfurane mediated dehydration of tertiary alcohols.

3.9 Asynchronous E2 anti elimination

This new tertiary alcohol dehydration method was exciting and such high diastereoselectivity cannot be explained by the commonly accepted E1 mechanism. Therefore, density functional calculations (Gaussian 09) were performed by Professor Daniel L. Ess, Yu Cai and Benjamin M. Kay to probe possible reaction pathways (E1, E2 and E1cb). The M06-2X/6-31+G(d,p) level of theory was chosen for its accuracy in other E2 transition state barrier calculations. Calculation shows that the E2-anti dehydration transition state has lower energy than the E1 carbocation intermediate and E2-syn dehydration transition state, and no E1cb transition state or carbanion intermediate was able to be located. These facts indicate that the dehydration proceeded via a highly asynchronous E2-anti transition state.1

3.10 Recourse to the transamidation

With this powerful dehydration method in hand and the experience we gained during the previous study, we reconsidered the transamidation strategy on ethyl ester 104 or 105, which
could be a most convenient transformation to incorporate ΔAAs into peptides. Recently, Costa and co-workers reported a DBU-catalyzed transamidation method from unactivated esters to amides.\textsuperscript{16} Encouraged by their results, we used ester 104 to test the conditions, however, no conversion was observed under reported conditions (Scheme 3.9).

\textbf{Scheme 3.9} Transamidation of ethyl ester 104.

Pirrung and coworkers reported a transamidation employing ethanolamine via an esterification–O→N acyl transfer sequence and they proposed a novel mechanism.\textsuperscript{17} The intramolecular hydrogen bonding between the amine and the alcohol of the ethanolamine makes the oxygen atom a better nucleophile to attack the mildly activated ester, effecting a transesterification process. A subsequent O→N acyl transfer would be driven by the formation of the more stable amide bond. To test this new chemistry, alkene 109 was produced via alkylation esterification–dehydration sequence with >20:1 dr from ester 80.

\textbf{Scheme 3.10} Synthesis of 2,2,2-trifluoroethyl ester 109.

2,2,2-Trifluoroethyl ester 109 was subjected to the ethanolamine–toluene conditions, but no conversion of the ester 109 to amide 110 was detected (entry 1, Table 3.5). Using additives such as NaHCO\textsubscript{3}, K\textsubscript{2}CO\textsubscript{3}, CeCl\textsubscript{3}, or adjusting reaction conditions, such as switching to other solvent systems or increasing loading of the ethanolamine were fruitless (entries 2–5, 7–9).
Similar to previous observations, the transamidation occurred while excess ethanolamine (21 equiv, entry 6, Table 3.5) was loaded into the system, delivering 110 with scrambled alkene stereochemistry as evidenced by NMR analysis of crude reaction mixtures. A reasonable explanation is that the large amount of amine promoted reversible conjugate addition to the alkene, thereby eroding the alkene integrity. Another possibility is that the basic conditions could promote azlactone formation, then subsequent ring opening delivered the product as alkene isomers. The ethanolamine–K$_2$CO$_3$–DMA combination also provided the transamidation product upon heating, and the alkene integrity was scrambled even with usage of 1.0 eq base, showing the challenge of performing such transformations on this delicate ester. Those observations excluded the application of this strategy to backbone protection-free ΔIle synthesis.

### Table 3.5 Investigation of the hydrogen bonding involved transamidation.

<table>
<thead>
<tr>
<th>entry</th>
<th>conditions</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.0 eq, Toluene, rt or 70 °C, 15 h</td>
<td>No reaction</td>
</tr>
<tr>
<td>2</td>
<td>3.0 eq 106, NaHCO$_3$, toluene, 70 °C, 15 h</td>
<td>No reaction</td>
</tr>
<tr>
<td>3</td>
<td>3.0 eq 106, NaHCO$_3$, THF–H$_2$O, rt, 60 h</td>
<td>No reaction</td>
</tr>
<tr>
<td>4</td>
<td>1.0–6.0 eq 106, Benzene, rt</td>
<td>No reaction</td>
</tr>
<tr>
<td>5</td>
<td>6.0 eq 106, acetic acid, cyclohexane, rt</td>
<td>No reaction</td>
</tr>
<tr>
<td>6</td>
<td>21.0 eq 106, cyclohexane, acetic acid, rt</td>
<td><strong>110</strong> with scrambled alkene configuration</td>
</tr>
<tr>
<td>7</td>
<td>1.5 eq 106, toluene, 50 °C, 1.5 h</td>
<td>No reaction</td>
</tr>
<tr>
<td>8</td>
<td>1.0 eq 106, CeCl$_3$, CH$_2$Cl$_2$, rt</td>
<td>Decomposition</td>
</tr>
<tr>
<td>9</td>
<td>1.0 eq 106, K$_2$CO$_3$, DMA, 60 °C</td>
<td><strong>110</strong> with scrambled alkene configuration</td>
</tr>
</tbody>
</table>
3.11 Advanced ester dehydration and O→N acyl transfer investigation

The lesson we learned from the previous ΔIle-containing peptide synthesis is that intermolecular couplings or transamidation processes cannot compete with the intramolecular azlactone formation (or possible intermolecular conjugate addition in certain cases). Either undesired pathway could cause the scrambling of the alkene integrity of ΔIle. To circumvent the azlactone formation, an activated ester should not be involved in the coupling process. However, to trigger an intermolecular transamidation to an unactivated ester, extra equivalents of free amine were required, and this in turn might promote the fatal reversible conjugate addition. Therefore, an intramolecular version of the transamidation, namely “O→N acyl transfer”, was adapted and corresponding model compound 111 was synthesized (Scheme 3.11). The phthalimide acted as a masked amine, which could be deprotected to reveal the free amine. We hypothesized that O→N acyl transfer could be triggered to deliver the more stable amide bond under appropriate conditions, and hopefully this activated-ester-free intramolecular transamidation would not scramble the alkene configuration. Starting from ester 77, the three-step sequence of saponification, alkylative esterification with glycine surrogate 78, and Martin sulfurane dehydration proceeded smoothly and delivered ester 111 in >19:1 dr (Scheme 3.11). The glycine surrogate 78 represented the least hindered amino acid connected to the C-terminus of an ΔIle residue in yaku’amide A. The facile dehydration of C-terminal β-hydroxylamino acids during couplings was frequently observed in our previous studies, which necessitated this alkylative esterification step. Unfortunately, decomposition of the starting material occurred inevitably during the following phthalimide cleavage step.
Possibly, the unsaturated ester moiety of 111 cannot survive the basic hydrazine conditions, which are commonly used to cleave phthalimide.

Scheme 3.11 Investigation of O→N acyl transfer using NPhth as amine surrogate.

With the failure of Phthalimide deprotection as well as the success of dehydrating tertiary alcohol 111, a new β-azido iodide 78a was used as an alternative in the alkylating step and delivered the azido ester 114 (Scheme 3.12). This choice was based on the facile reduction of azides under mild Staudinger reduction conditions. Gratifyingly, the alkylation and Martin sulfurane mediated dehydration delivered alkene 115 with excellent results. Also, the crucial azide reduction proceeded smoothly with 3 equivalents of PMe₃ at 0 ºC. However, careful analysis of the crude NMR obtained after aqueous workup as well as the TLC (Rₜ value 0.0 under 10% MeOH/CH₂Cl₂ conditions, being sensitive to the Ninhydrin stain) disclosed that the ester 116 did not spontaneously rearrange to form the respective amide 117 (Table 3.6) automatically. Thus, further manipulation was required to trigger the O→N transfer process.
Scheme 3.12 Investigation of O→N acyl transfer using azide as amine surrogate.

To convert the amine 116 to amide 117, the solvent effect was considered at first. Various solvent systems were screened but in vain (entries 2–8, Table 3.6). The mild base pyridine could not promote the acyl transfer chemistry either (entry 6). Fortunately NaHCO₃ in THF–H₂O promoted the acyl transfer chemistry and delivered the corresponding amide 117 in excellent yield and 6:1–9:1 dr (entry 1). In addition, we found that various organic bases could trigger the acyl transfer chemistry with different rates (entries 10–15), while some bases led to decomposition (entry 16) or isomerization (entry 17). We also observed that a reverse “N→O acyl transfer” could happen to amide 117 under acidic conditions. Ultimately, the use of morphline or piperidine afforded the ΔIle-containing peptide 117 in >10:1 dr (entries 10 and 13).
Table 3.6 Testing of the O→N acyl transfer process.

![Reaction diagram]

<table>
<thead>
<tr>
<th>entry</th>
<th>condition\textsuperscript{a}</th>
<th>result\textsuperscript{b}</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>NaHCO\textsubscript{3}, THF–H\textsubscript{2}O, 24 h</td>
<td>80%, 6:1–9:1 dr</td>
</tr>
<tr>
<td>2</td>
<td>Toluene, 24–168 h:</td>
<td>No progress</td>
</tr>
<tr>
<td>3</td>
<td>THF, 24–168 h</td>
<td>No progress</td>
</tr>
<tr>
<td>4</td>
<td>THF, 50 °C, 14 h</td>
<td>Sluggish progress with isomerization</td>
</tr>
<tr>
<td>5</td>
<td>CDCl\textsubscript{3}, 408 h</td>
<td>Half conversion to 117</td>
</tr>
<tr>
<td>6</td>
<td>Benzene, 72 h</td>
<td>Sluggish progress</td>
</tr>
<tr>
<td>7</td>
<td>Pyr, 24–72 h</td>
<td>No progress</td>
</tr>
<tr>
<td>8</td>
<td>Toluene, 35 °C, 15 h</td>
<td>Sluggish progress with isomerization</td>
</tr>
<tr>
<td>9</td>
<td>Toluene, 50 °C, 14 h</td>
<td>Alkene isomerization</td>
</tr>
<tr>
<td>10</td>
<td>Silical gel, THF, 24 h</td>
<td>No progress</td>
</tr>
<tr>
<td>11</td>
<td>Piperidine, THF–H\textsubscript{2}O, 24 h</td>
<td>67%, 11.5:1 dr</td>
</tr>
<tr>
<td>12</td>
<td>Piperidine, EtOAc–H\textsubscript{2}O</td>
<td>Very sluggish progress</td>
</tr>
<tr>
<td>13</td>
<td>N-methylmorpholine, THF–H\textsubscript{2}O, 60 h</td>
<td>Sluggish progress</td>
</tr>
<tr>
<td>14</td>
<td>Morpholine, THF–H\textsubscript{2}O, 60 h</td>
<td>60%–90%, 10.5:1–16:1 dr</td>
</tr>
<tr>
<td>15</td>
<td>Morpholine, THF, MS 4Å, 15 h</td>
<td>Sluggish progress with decomposition</td>
</tr>
<tr>
<td>16</td>
<td>Morpholine, THF–H\textsubscript{2}O, 40 °C, 15 h</td>
<td>60%, 6:1 dr</td>
</tr>
<tr>
<td>17</td>
<td>NEt\textsubscript{3}, 48 h</td>
<td>Possible decomposition</td>
</tr>
<tr>
<td>18</td>
<td>\textit{N,N,N\textprime} Trimethylsilyldiamine, THF–H\textsubscript{2}O, 24 h</td>
<td>2:1 dr</td>
</tr>
</tbody>
</table>

\textsuperscript{a} All reactions were performed at rt, unless marked with other temperature.

\textsuperscript{b} The \textit{dr} is the intergration ratio in NMR between Z-\textDelta Ile product and \textit{E}-\textDelta Ile side product.

3.12 The synthesis of model peptide Cbz-Gly-Z-\textDelta Ile-D-Valinol

The success of the synthesis of Cbz-Gly-Z-\textDelta Ile-Glycinol 117 was exciting and encouraged us to test the feasibility of this method with other \textDelta Ile-containing model peptides. Very similarly, the ester 119 was obtained efficiently from ester 77 (Scheme 3.13). Here, the valine surrogate 78b represented the most hindered amino acid connected to the \textit{C}-terminus of \textDelta Ile residue in yaku’amide A, which was assumed to be the most challenging substructure. The Martin
sulfurane dehydration happened smoothly with excellent yield and dr. Upon Staudinger reduction using PMe₃, the azide 119 was reduced to amine, and one-pot addition of base triggered the O→N acyl transfer and delivered amide 120 with minimal alkene isomerization. Interestingly, we observed the dr of the products from the O→N acyl transfer varied under THF–H₂O–morpholine and DMF–H₂O–piperidine conditions. Nonetheless, both combinations delivered usable results to be applied to total synthesis.

![Scheme 3.13](image.png)

Note: Stereocenters marked by asterisks possess the indicated relative stereochemistry. (This rule works for all the chapters of this dissertation.)

**Scheme 3.13** Synthesis of the Cbz-Gly-Z-ΔIle-D-Valinol.

### 3.13 The synthesis of E-ΔIle-containing model peptides

In the following stage, we were curious to test the diastereoselectivity after the O→N acyl transfer reaction on E-ΔIle residue-containing peptides. Gratifyingly, this saponification–alkylative esterification–anti dehydration sequence worked equally well as the previous Z-ΔIle related synthetic sequence (Schemes 3.12 and 3.13). Isomerically pure alkenes 122a and 122b were obtained with high yields and subjected to Staudinger reduction and base-catalyzed
rearrangement conditions (Scheme 3.13). The Z-Δlle-containing tripeptides 110 and 123 were obtained with high efficiency and minimal E-to-Z isomerization (>10:1 dr).

Scheme 3.14 Synthesis of the E-Δlle-containing model peptides.

3.14 The synthesis of Z(E)-Δlle-Alaninol-containing model peptides

With the successful incorporation of the least and most sterically hindered amino acids at the C-termini of the E- and Z-Δlle-containing dipeptides, we tested the feasibility of this strategy to install an alanine residue to the C-terminus of Δlle, which is also present in yaku’amide A. Surprisingly, different reactivity was observed during the course of incorporating this medium-sized residue (Scheme 3.15). The first variation happened in the alkylative esterification process: the standard β-azido iodide (78c)–NEt₃ conditions only delivered 30–60% yield after purification, instead of the 80–90% for the glycine and valine surrogates. The second variation was observed during the O→N acyl transfer process: 4:1–6:1 dr was obtained consistently. These interesting results are unusual compared to the results obtained during the synthesis of Gly and Val-containing substrates, which indicated that optimization of reaction conditions in the following total synthesis might be required. Unfortunately, we cannot find any compelling arguments to
explain this phenomenon except that the reactivity difference was caused by subtle structural changes.

Scheme 3.15 Synthesis of the Cbz-Gly-\(Z(E)\)-\(\Delta\)Ile-\(D\)-Alaninol.

3.15 Conclusion

We have developed a method of incorporating \(E\)- or \(Z\)-dehydroisoleucine(\(\Delta\)Ile) residues into peptides via a Martin sulfurane dehydration–Staudinger reduction–\(O\rightarrow N\) acyl transfer sequence. The key transformation is an unusual Martin sulfurane mediated \textit{anti} dehydration of \(\beta\)-OHAA derivatives. This highly efficient method did not require the backbone amide protection chemistry, which is necessary in all other known routes. The success of the model study set the stage for initiating the total synthesis of yaku’amide A.
3.16 References


Chapter 4.  PROGRESS TOWARDS THE SYNTHESIS OF YAKU’AMIDE A

With the success of the model study to deliver ΔIle-containing dipeptides, we initiated the total synthesis of yaku’amide A’.\(^1\)

4.1 Retrosynthetic analysis

Our retrosynthesis of yaku’amide A’ was designed to maximize synthetic convergence based on our O→N acyl transfer chemistry (Chapter 3). Retrosynthetically, 1 is disconnected at two key amide bonds producing three subunits: the \(N\)-terminus 130, left-hand pentapeptide 131 and right-hand nonapeptide 132 (Scheme 4.1). The specific protecting groups (PGs) would be identified through experimentation.\(^2\) In the long run, we plan to synthesize the \(N\)-terminal acyl group via Myer’s alkylation\(^3\) followed by a Mukaiyama-type aldol reaction.\(^4\) Inoue’s method,\(^5\) although lengthy, would be suitable for quick access to the \(N\)-terminal acyl group. The construction of the left-hand pentapeptide 131, containing a β-OH\(\text{Ile}-\text{Z-\text{Ile-Gly-E-\Delta-Ile-Val}}\) sequence, demands a linear synthesis as a result of two dehydration–azide reduction–O→N acyl transfer sequences. To access nonapeptide 132, there are two possible key scissions at the peptide backbone, which lead to the routes \(L\) and \(R\).

Further scission of those key subunits revealed several smaller intermediates. Scission of pentapeptide 131 revealed tetrapeptide 133 and valine surrogate 78b. Peptide 133 was further divided into tripeptide 134 and racemic β-OH isoleucine 135. An alkylation–dehydration–azide
reduction–O→N acyl transfer sequence can fuse dipeptide 136 and glycine surrogate 78a together to deliver tripeptide 134, and the C-terminus primary alcohol can be oxidized prior to coupling with racemic aminohydroxylation adduct 135 (synthesized in Chapter 3). Dipeptide 136 can be accessed via coupling of the optically active β-OHlle derivative with the racemic aminohydroxylation adduct. The synthesis of subunit 131 would cost most linear steps due to the presence of two ∆Ile residues, and we expected it to be the most challenging part of the project.

To access nonapeptide 132 via route L, scission of the amide bond connecting the two β-OH-Val residues revealed the dipeptide 137 and heptapeptide 138, which can be further divided into ∆Ile-containing tripeptide 139 and C-terminal tetrapeptide 140 (Scheme 4.1). Peptide 139 can be obtained via a similar strategy to the one proposed for tripeptide 134. Specifically, alkylation product from dipeptide 141 with alanine surrogate 78c followed by the dehydration–azide reduction–O→N acyl transfer sequence can afford 139. To synthesize the C-terminal tetrapeptide 140, the ∆Val is the key fragment. Based on the observation of EDCI·HCl-promoted azlactone formation in our lab (vide infra), we proposed to apply the azlactone formation–azlactone ring opening chemistry to construct 140, which enables the scission of C-terminus of ∆Val to deliver 142 and known amine 143. The tripeptide 142 can be provided straightforwardly via coupling of the dipeptide 144 and racemic aminohydroxylation adduct 145.

Route R represents another possible means of constructing nonapeptide 132 as well. Fragments 146 and 140 are delivered from the first key scission of 132. Further scission of 146 reveals tetrapeptide 147 and alanine surrogate 78c, and the former could be accessed via coupling of dipeptides 148 and 149.
Scheme 4.1 Retrosynthetic Analysis.
4.2 Synthesis of the right-hand nonapeptide

In targeting the natural product, we chose the right-hand nonapeptide 132 (Scheme 4.1) as the first synthetic goal since it could be prepared in fewer linear steps than the left-hand pentapeptide 131. Only one ΔIle residue is incorporated in 132, rendering it easier to access than 131.

4.2.1 Investigation of route R

Of the two possible synthetic routes towards 132 (Scheme 4.1), we initially examined route R, which is more convergent. In addition, route R will require one less chromatographic separation of tertiary amine-containing intermediates than route L, and the purification of such compounds amines can be difficult and time-consuming.

4.2.1.1 Synthesis of enantio-rich D- and L-β-OH Vals

To access the β-OH amino acids enantioselectively, co-workers J. Jiang and J. M. Cardon had thoroughly investigated the asymmetric oxyamination. However, useful levels of enantioselectivity were not observed. Fortunately, they discovered a convenient alternative to access enantiomerically enriched β-OHAAAs via aminohydroxylation employing Lebel’s chiral mesyloxycarbamate 2 (Scheme 4.2). The two diastereomers 148 (1:1 dr) generated via aminohydroxylation, were protected as silyl ethers, and then separated on silica gel. Three chromatographic separations were required to deliver 100 mg of each isomer (dr ≥10:1) from 1 g of the mixture (1:1 dr), providing sufficient material for testing synthetic routes. The attempts to increase the separation efficiency via switching the silyl protecting group were fruitless. Similarly, we can access the desired (2S,3R)-β-OHlle using the same sequence. This
advantageous method enables production of both required β-OHVal isomers in two steps with the required functional groups protected.

![Scheme 4.2 Aminohydroxylation employing Lebel’s carbamate.](image)

### 4.2.1.2 Synthesis of β-OH containing dipeptides

With success in accessing enatioenriched β-hydroxy amino acids, we began the synthesis of dipeptide 154. Attempted saponification of ethyl ester 149 under traditional methods was problematic. The common basic hydrolysis conditions (e.g., LiOH, NaOH, KOH) inevitably led to the cleavage of the chiral carbamate residue. In light of Nicolau’s work on mild hydrolysis of methyl esters using SnMe₃OH, we adapted these conditions and delivered the desired carboxylic acid from 149. However, subsequent coupling to the racemic amine 153 only afforded 20–47% yield from ester 149, which may be caused by the inefficient hydrolysis of the robust ethyl ester. This method also suffers from a common disadvantage: ten or more equivalents loading of the toxic and expensive SnMe₃OH is required to promote full conversion. Sufficient material was provided via this inefficient two-step protocol, which enabled the testing of subsequent reactions.
Scheme 4.3 Synthesis of dipeptide 155.

To access the coupling partner of 155, the chiral auxiliary of the ester 150 was reductively cleaved under Zn-AcOH conditions and coupling to Boc-D-allo-isoleucine delivered the dipeptide 154 in 77% yield (Scheme 4.4). The Zn–Acetic acid conditions require chromatographic purification to deliver the amine product after Celite filtration, which is inconvenient. The vulnerability of chiral auxiliary observed in the ethyl ester saponification process inspired us to test the LiOH–H₂O–t-BuOH conditions for cleavage of the chiral auxiliary. Disappointingly, although the amine product was present in mass spectrometry, following coupling to Boc-D-allo-isoleucine was not able to provide 156 with useful yields. Possibly, this low yielding transformation was the consequence of inefficient aqueous workup or base-promoted decomposition of the starting material. Therefore, this method was abandoned and the Zn-AcOH conditions were set as the standard chiral auxiliary cleavage conditions. Next, the LiOH-promoted hydrolysis revealed the free carboxylic acid 157 without incident.
Scheme 4.4 Synthesis of dipeptide 157.

4.2.1.3 Synthesis of central pentapeptide

With the two advanced dipeptides 155 and 157 in hand, we tested the coupling reaction to deliver the tetrapeptide 158. EDCI–HOAt conditions were tested first due to their success in the synthesis of dipeptide 156 (Table 4.1). Surprisingly, this coupling reaction was extremely challenging. Attempts to tune the bases in EDCI–HOAT conditions were fruitless (entries 2–4). COMU–2,4,6-collidine conditions failed as well (entry 5). The DEPBT–NEt3 combinations delivered milligram quantities of the product in 20% yield with concomitant decomposition (entry 1). The difficulty of this coupling may be caused by the bulkiness of the two tert-butyldimethylsilyl (TBS) protecting groups.

Table 4.1: Study of the tetrapeptide synthesis.

<table>
<thead>
<tr>
<th>entry</th>
<th>conditions</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>DEPBT, NEt3, THF–DMF</td>
<td>20% with Decomposition (Not reproducible)</td>
</tr>
<tr>
<td>2</td>
<td>HOAt, EDCI, THF–DMF</td>
<td>Decomposition</td>
</tr>
<tr>
<td>3</td>
<td>HOAt, EDCI, Hunig’s base, THF–DMF</td>
<td>Decomposition</td>
</tr>
<tr>
<td>4</td>
<td>HOAt, EDCI, Na2CO3, THF–DMF</td>
<td>Decomposition</td>
</tr>
<tr>
<td>5</td>
<td>COMU, 2,4,6-collidine, NEt3, THF–DMF</td>
<td>Decomposition</td>
</tr>
</tbody>
</table>
4.2.1.4 Testing of the O→N acyl transfer chemistry

With this precious tetrapeptide 158 in hand, we proceeded forward and tested the following reactions. The saponification–alkylative esterification progressed smoothly at 75% yield, and the following Martin sulfurane mediated dehydration delivered the required alkene 160 at excellent yield without observation of the other alkene isomer (Scheme 4.5). This advance set the stage to test the azide reduction–O→N acyl transfer chemistry. Azide reduction promoted by Lindlar catalyst (Sigma-Aldrich)–H₂ happened smoothly, and one-pot addition of piperidine delivered the pentapeptide in 80% yield. Careful analysis of the reaction mixture revealed that no isomerized product was detected, showing the feasibility of this chemistry with complicated intermediates. In this sequence of reactions, several modifications of the chemistry used in the model study were explored with good results. Cs₂CO₃ exhibited superior activity than the NEt₃ in the alkylation step and afforded excellent conversion, and the Lindlar catalyst–H₂ conditions successfully suppressed the isomerization possibly caused by the conjugate addition of PMe₃.

Scheme 4.5 Test of the O→N acyl transfer chemistry.
Since the bulkiness of the tert-OTBS were presumably hindering the tetrapeptide coupling reaction, we were concerned that their cleavage might be problematic. Therefore, the cleavage of tert-TBS were tested on a mixture of 149 and 150 (1:1 dr). Unfortunately, the common TBAF or HF·Pyr conditions did not promote silyl cleavage (Scheme 4.6). We were hesitant to apply stronger conditions, which might cause epimerization or decomposition of later stage intermediates. Afterwards, the less robust protecting group triethysilyl (–TES) was chosen to replace the tert-TBS group. Starting with the –TES-protected amino acids 151 and 152 (Scheme 4.2), we attempted to perform the route R but the synthesis of the respective tetrapeptide was problematic again. Finally, we abandoned route R.

![Scheme 4.6 Attempts of TBS group cleavage.](image)

4.2.2 Investigation of route L

In the course of investigating route R, we gained valuable experience and lessons for the peptide coupling as well as the functional group transformations. With the failure of route R, we moved forward to investigate route L.

4.2.2.1 Optimization of the Me₃SnOH hydrolysis reaction

We reasoned the problematic hydrolysis–coupling sequence en route to 154 (Scheme 4.3). The inefficient hydrolysis required the use of large amount of Me₃SnOH, which was hard to completely remove via the acidic aqueous workup. The remaining Me₃SnOH or its derivative may be the culprit decreasing the efficiency of the coupling reaction. Since a large amount of
white precipitate was observed during the hydrolysis process in the standard (CH$_2$Cl)$_2$ media, we hypothesized that the poor solubility of Me$_3$SnOH possibly caused the inefficient transformation. Therefore, we finely tuned the parameters of hydrolysis of ester 151 (Scheme 4.7) by testing various solvents, gradually decreasing the Me$_3$SnOH loadings, and adjusting the reaction temperatures. Ultimately, we were pleased to find that 2–3 eq Me$_3$SnOH in hexane at 60 °C is the optimum condition, representing the lowest Me$_3$SnOH loading that can still furnish full conversion of starting material. This choice of solvent took advantage of the low polarity of our chiral ester 151. In addition, to overcome the shortcomings of acidic aqueous workup (especially since the –TES protecting group is sensitive to acidic conditions), a Celite pad filtration of the reaction mixture followed by diethyl ether wash delivered the desired carboxylic acid 152 with high purity, which was used in the coupling reaction without chromatographic purification. To the best of our knowledge, this is the first time that the hexane–SnMe$_3$OH combination was observed to promote ethyl ester hydrolysis with extremely low SnMe$_3$OH loadings. This novel Celite filtration–workup can be employed to handle acid- or base-sensitive substrates.

![Scheme 4.7 Hydrolysis of the ester 151.](image)

4.2.2.2 Optimization of the dipeptide coupling

With high quality product from the hydrolysis reaction, the following coupling reaction was investigated and it turned out quite eventful. The TES group cleavage was often observed in the standard EDCI–HOBt coupling conditions (Table 4.2). Therefore, a detailed study was carried out to overcome this drawback, using the mass spectrometry to monitor the TES cleavage.$^{10}$
Adjusting the loading of coupling reagents, switching the solvent from THF–DMF mixture to pure THF or DMF and varying the reaction time did not produce positive results (entries 1 and 3). Using NaHCO$_3$ as an additive directly led to the TES group cleavage (entry 2). Surprisingly, the DCC–HOBt–CH$_2$Cl$_2$ conditions gave superior results and 164 was the only detected product (entry 4). However, while DCC–HOBt were combined with THF or THF–DMF solvent system, the –TES cleavage product 164a was observed again (entries 5–6). After careful analysis of those conditions, we concluded that the solvent might be the key to this coupling reaction. Rapidly, EDCI–HOBt–CH$_2$Cl$_2$ was tested and delivered 164 as single product with >80% yield with good reproducibility (entry 7). Due to difficulties in the removal of DCU side product from the reaction mixture (entry 4), we chose EDCI·HCl as the coupling reagent (entry 7).

![Chemical structures](image)

<table>
<thead>
<tr>
<th>entry</th>
<th>coupling condition</th>
<th>result (the ratio of 164:164a in MS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>EDCI, HOBt, THF–DMF, 3–15 h</td>
<td>Inconsistent, varies from 3:1 to 164 as single product</td>
</tr>
<tr>
<td>2</td>
<td>EDCI, HOBt, NaHCO$_3$, THF–DMF</td>
<td>Only 164a</td>
</tr>
<tr>
<td>3</td>
<td>EDCI, HOBt, THF or DMF</td>
<td>Inconsistent, varies from 3:1 to 164 as single product</td>
</tr>
<tr>
<td>4</td>
<td>DCC, HOBt, CH$_2$Cl$_2$, 3 h</td>
<td>Only 164, ca. 80%</td>
</tr>
<tr>
<td>5</td>
<td>THF, DCC, HOBt, 2.5 h</td>
<td>1.5:1</td>
</tr>
<tr>
<td>6</td>
<td>DCC, HOBt, THF–DMF, 15 h</td>
<td>Inconsistent, 164a or 164 as the only product</td>
</tr>
<tr>
<td>7</td>
<td>EDCI, HOBt, CH$_2$Cl$_2$, 2 h</td>
<td>Only 164, 83%, reproducible</td>
</tr>
</tbody>
</table>

Table 4.2 Optimization of dipeptide coupling.

4.2.2.3 **Protecting group switch and alkylation**

With the puzzle of dipeptide 164 solved, the following hydrolysis and alkylation sequence was tested. In contrast to the route R study, this sequence was quite eventful. Due to the liability towards inorganic bases, partial cleavage of the chiral auxiliary of 164 was consistently observed
during the hydrolysis and alkylation steps. Significant effort was taken to optimize this reaction, such as adjusting the temperatures, the amount of base, and changing solvents, but no positive result was obtained. Interestingly, dehydrovaline-like structural features were shown by NMR analysis of the crude mixture.

Scheme 4.8 Attempts to synthesis dipeptide with alanine surrogate.

Next, we considered using the less efficient strategy of switching the chiral auxiliary into a base-stable protecting group, which would add extra steps to the total synthesis. The tert-butylloxycarbonyl (Boc) group was chosen due to its robustness towards basic conditions as well as facile removal, and hopefully, its removal could happen concomitantly with the TES group cleavage. To maintain the efficiency of our synthesis, a one-pot conversion was investigated to effect the cleavage of the chiral auxiliary as well as installation of the Boc protecting group to the resulting amine intermediate. As expected, cleavage of the fragile TES group happened during the weakly acidic reaction conditions (Pd/C, Boc₂O without adding base, Table 4.3, entry 1). Attempts to use Na₂CO₃ or NaHCO₃ as basic additives were also not effective (entries 2 and 3). Employing a one-pot, 2-step conversion via cleaving the chiral auxiliary then adding Boc₂O–NEt₃ was unsuccessful (entry 4), and analysis of the amine intermediate obtained in the first step demonstrated the hydrogenolysis conditions can cause TES cleavage as well. Recently, Sajiki, Hirota and co-workers reported the advantage of using CH₃CN as the hydrogenolysis solvent in retaining the TES protecting group.¹¹ Unfortunately, CH₃CN did not bring any improvement in
suppressing TES cleavage. Ultimately, we concluded that the inorganic basic additives in the organic solvent may not be effectively quenching the acids due to their low solubility. As expected, using saturated aqueous NaHCO₃ as the cosolvent successfully delivered the desired product without detectable TES cleavage. Upon optimization, 100 Psi H₂, THF–Sat. aq. NaHCO₃ (V_{THF} : V_{Sat. aq. NaHCO₃} = 3:1) was used in the reaction to effect rapid transformation. Also, we used aqueous extraction as the workup instead of Celite filtration since H₂O was present in the reaction mixture.

**Table 4.3** Investigation of one-pot transformation of Chiral auxiliary→Boc.

<table>
<thead>
<tr>
<th>entry</th>
<th>condition</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Boc₂O, MeOH, 60 Psi H₂</td>
<td>Partial TES group cleavage observed</td>
</tr>
<tr>
<td>2</td>
<td>Boc₂O, MeOH, NaHCO₃, 60 Psi H₂</td>
<td>Partial TES group cleavage observed</td>
</tr>
<tr>
<td>3</td>
<td>Boc₂O, MeOH, Na₂CO₃, 60 Psi H₂</td>
<td>Partial TES group cleavage observed</td>
</tr>
<tr>
<td>4</td>
<td>Boc₂O, EtOH, 60 Psi H₂; then Boc₂O, NEt₃</td>
<td>Partial TES group cleavage observed</td>
</tr>
<tr>
<td>5</td>
<td>Boc₂O, THF, NaHCO₃, 60 Psi H₂</td>
<td>Partial TES group cleavage observed</td>
</tr>
<tr>
<td>6</td>
<td>Boc₂O, CH₃CN, 60 Psi H₂</td>
<td>Partial TES group cleavage observed</td>
</tr>
<tr>
<td>7</td>
<td><strong>THF–Sat. NaHCO₃, 3:1, 100 Psi H₂, 15 h</strong></td>
<td>166, &gt; 90% isolated yield, reproducible</td>
</tr>
</tbody>
</table>

With success in obtaining Boc-protected dipeptide 166, the LiOH-promoted saponification happened smoothly and delivered the corresponding carboxylic acid 167 (Table 4.4). The alkylation conditions (iodide, DMF, NEt₃, 80 °C) developed in the model study were sluggish and did not achieve full conversion (entry 1). Additives and different solvents were not effective (Scheme 4.4, entries 2–6). During the process of optimization, we found the reactions with only “partial conversion” observed in MS always led to a low yield (<40%). This correlation made mass spectrometry a convenient tool to monitor the alkylation progress. Other bases, such as
Na₂CO₃, K₂CO₃, did not afford good conversion either (entries 8–9). The Cs₂CO₃–DMF system was most promising, and careful control of loading is crucial. The small scale testing reactions made the strict loading of Cs₂CO₃ difficult, causing variations in the yields (entries 10–15). Especially, extra loading of Cs₂CO₃ could lead to the generation of only retroaldol product. However, the minimum 50% yield is sufficient to move the synthesis forward.

Table 4.4 Synthesis of advanced ester 168 and optimization of the alkylation.

<table>
<thead>
<tr>
<th>entry</th>
<th>condition</th>
<th>result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.0 eq NEt₃, DMF, 60 °C, 15 h</td>
<td>Partial conversion</td>
</tr>
<tr>
<td>2</td>
<td>3.0 eq NEt₃, 4 eq AgNO₃, 70°C, 7 h</td>
<td>Partial conversion with retroaldol product</td>
</tr>
<tr>
<td>3</td>
<td>1.0 eq NEt₃, DMA, 70 °C, 15 h</td>
<td>Partial conversion</td>
</tr>
<tr>
<td>4</td>
<td>1.0 eq NEt₃, CH₂Cl₂, 70 °C, 20 h</td>
<td>Partial conversion</td>
</tr>
<tr>
<td>5</td>
<td>1.0 eq NEt₃, Hexane, 60 °C, 15 h</td>
<td>No reaction</td>
</tr>
<tr>
<td>6</td>
<td>1.0 eq NEt₃, DMF, 20 h</td>
<td>Partial conversion</td>
</tr>
<tr>
<td>7</td>
<td>1.0 eq NEt₃, DMSO,70 °C, 20 h</td>
<td>Partial conversion</td>
</tr>
<tr>
<td>8</td>
<td>2.0 eq Na₂CO₃,50 °C, 15 h</td>
<td>Partial conversion with retroaldol product</td>
</tr>
<tr>
<td>9</td>
<td>1.5 eq K₂CO₃, 18-crown-6, DMF, 50 °C, 15 h</td>
<td>All retroaldol product with 167.</td>
</tr>
<tr>
<td>10</td>
<td>1.0 eq CsCO₃, DMF, 60 °C, 15 h</td>
<td>87% with minor retroaldol</td>
</tr>
<tr>
<td>11</td>
<td>0.5 eq CsCO₃, DMF, 75 °C, 15 h</td>
<td>Partial conversion</td>
</tr>
<tr>
<td>12</td>
<td>0.75 eq Cs₂CO₃, DMF, 75 °C, 15 h</td>
<td>Mostly converted to 168 with minor 167</td>
</tr>
<tr>
<td>13</td>
<td>1.04 eq Cs₂CO₃, DMF, 60 °C, 15 h</td>
<td>50%–87%</td>
</tr>
<tr>
<td>14</td>
<td>1.1 eq Cs₂CO₃, dioxane, 70 °C, 15 h</td>
<td>Partial conversion</td>
</tr>
<tr>
<td>15</td>
<td>1.5 eq Cs₂CO₃, DMF, 60 °C, 15 h</td>
<td>58%–88%</td>
</tr>
</tbody>
</table>

4.2.2.4 Furnishing the tripeptide

The successful synthesis of the advanced ester 168 set the stage to perform the dehydration–azide reduction–O→N acyl transfer sequence. Upon treatment of alcohol 168 with
Martin sulfurane, excellent yield and diastereoselectivity were obtained without incident (Scheme 4.9). Considering the possibility that the alkene isomerization was caused by conjugate addition by the basic PMe₃, Lindlar catalyst–H₂ conditions were chosen to reduce the azide and it worked smoothly. Then, one-pot addition of piperidine triggered the O→N acyl transfer with excellent results. For the dehydration step, separation of the alkene product from the diphenyl sulfoxide side product on silica gel was difficult due to the similar polarity on silica gel. Diphenyl sulfoxide was derived from Martin sulfurane. Since there was no apparent conflict between the diphenyl sulfoxide side product and subsequent reaction conditions, we proposed the more convenient one-pot three-step transformation: the solvent of the crude dehydration mixture could be evaporated and H₂–Lindlar catalyst–THF–H₂O would be added to the same vial. Upon completion of azide reduction, base could be added to trigger the O→N acyl transfer chemistry. Gratifyingly, the first attempt of this reaction met with success. The azide was reduced to the corresponding amine smoothly, and addition of piperidine promoted the acyl transfer chemistry (Scheme 4.9). The tripeptide 169 was obtained via a facile chromatographic column separation in 75% yield and 12:1 dr. This set of reactions features the convenient performance of three chemical transformations in one vial without isolation of the reaction intermediates, and the high efficiency of this process was maintained.

**Scheme 4.9** One-pot 3-step transformation from ester to amide.
4.2.2.5 **Synthesis of the C-terminal tetrapeptide**

During the course of synthesizing activated esters (Chapter 3), EDCI–HOBT conditions were attempted to deliver the tert-Butyl ester, and this operation brought the unexpected discovery of the azlactone (oxazalone) 170 formation, possibly via EDCI•HCl promoted double dehydration. Upon further exploration, we found that the azlactone ring opening via attack of amine was widely used to install dehydroamino acids with symmetric alkenes, and dehydrovaline fit this category.\(^{12}\)

![Scheme 4.10 Discovery of azlactone formation promoted by EDCI-HCl.](image)

This result prompted us to take advantage of the facile azlactone formation to synthesize the right-hand tetrapeptide. Due to the symmetric feature of dehydrovaline, alkene isomerization is not a concern in this process. Therefore, the coupling of known dipeptide 144 to amine 145 (derived from an aminohydroxylation adduct) followed by LiOH-promoted hydrolysis delivered tripeptide 142. Upon treatment with 2 equiv EDCI at room temperature for 24 h, the azlactone intermediate was delivered (monitored via MS). Then, one-pot addition of the required amine•HCl salt 143 and NEt\(_3\) produced the N-Boc protected tetrapeptide in 90% yield within 3 h at room temperature. It is noteworthy that commonly used heating to promote the azlactone ring opening was not required in this process with the presence of extra organic base. Further treatment of 140 with HCl smoothly delivered amine•HCl salt 171 in quantitative yield.
4.2.2.6 Completion of the nonapeptide

In our model study, oxidation of the primary alcohol to the carboxylic acid was required for the peptide chain elongation. Among the tested conditions, TPAP-NMO conditions delivered low yields product with poor reproducibility and the TEMPO-bleach mediated one-step condition did not even consume the starting material. Finally, the one-pot two-step oxidation using Dess–Martin periodinane followed by Pinnick conditions delivered the carboxylic acid effectively in our model study. With all the required fragments prepared in the lab, we initiated the coupling reactions to fuse them together. Tripeptide 169 was smoothly oxidized using this DMP–Pinnick oxidation strategy, but the 2-Iodobenzoic acid byproduct interfered with the following coupling reaction with tetrapeptide 171 (Scheme 4.12). The 2-Iodobenzoic acid is derived from Dess–Martin periodinane and is always co-extracted out of the acidic aqueous phase with the acid 172. To remove this by product from acid 172 and avoid chromatographic operations, we took advantage of the low polarity of the carboxylic acid 172. Sat. aq. NaHCO$_3$ was added to basify the oxidation reaction mixture, and large volumes of EtOAc were used to extract the sodium salt of the carboxylic acid 172 out of the aqueous media. Pleasantly, we found most of the Iodobenzoic acid stayed in the aqueous media and the minor acid co-extracted could be removed by chromatographic separation after the coupling reaction. Coupling of 172 to 171
worked smoothly and delivered product 173 in 83% yield from alcohol 169. Then, HCl-promoted Boc cleavage delivered amine•HCl salt 174 with concomitant cleavage of the TES group. Evaporation of the HCl-ether mixture and coupling to the carboxylic acid 175 delivered the right-hand nonapeptide 176 in 56% yield. It is noteworthy that retroaldol side product was predominantly formed in this coupling reaction when THF or DMF were used as the solvent. Upon investigation, using CH$_2$Cl$_2$ as solvent successfully suppressed the retroaldol reaction.

![Scheme 4.12 Synthesis of right-hand nonapeptide 176.](image)

4.3 **Synthesis of left-hand pentapeptide**

With the successful right-hand nonapeptide synthesis, considering the similarity between tripeptide 169 and the left-hand pentapeptide, we initiated our synthesis using the same chemistry. Beginning with the aminohydroxylation of Z-enoate, followed by TES protection and
chromatographic separation, carbamate 177 was obtained, and was further submitted to the Me₃SnOH-promoted hydrolysis–coupling sequence (Scheme 4.13). The dipeptide 177 was obtained in 74% yield over two steps, which was quantitatively converted to the Boc-protected dipeptide 178. The saponification and standard alkylation conditions (developed in our model study) delivered advanced ester 179 without incident, setting the stage for the one-pot three-step peptide construction. The dehydration and azide reduction was evidenced by MS and TLC, and finally piperidine addition promoted the O→N acyl transfer and delivered the tripeptide 180 containing a primary alcohol. Afterwards, Dess-Martin periodane–Pinnick oxidation converted 180 into carboxylic acid 181, which was coupled to racemic amine 182 (derived from an aminohydroxylation adduct). The first attempt of the alkylation esterification–anti dehydration–azide reduction–O→N acyl transfer sequence was successful and left-hand pentapeptide 186 was obtained efficiently.
After significant advances in the synthesis of yaku’amide A' (Figure 1.1, Chapter 1) had been achieved in our lab, we were notified that the structure of yaku’amide A was revised. Therefore, no further effort was devoted to this dead end route, such as scaling up the synthesis of heptapeptide and nonapeptide and respective characterizations. Instead, we immediately initiated the synthesis of the nonapeptide with the correct configuration, and this nonapeptide was delivered within three weeks using the same chemistry.\textsuperscript{15} In addition, a microscale reaction testing the coupling of right-hand nonapeptide and left-hand pentapeptide was successful, and the formation of respective tetradecapeptide was well-evidenced by MS. This rapid re-synthesis process demonstrates the feasibility and robustness of our route towards the natural product.
4.5 Conclusion

Significant progress towards the synthesis of yaku’amide A has been achieved in the lab, such as the left-hand pentapeptide and the right-hand nonapeptide synthesis. In this process, the sequence of Martin sulfurane anti dehydration–azide reduction–O→N ayl transfer was able to be performed in one-pot and successfully applied to the synthesis of ΔIle-containing complicated peptide intermediates. With all the advance in this project, we are confident to access yaku’amide A in the near future.

4.6 References

(1) Since the structure revision was reported very recently, the beginning retrosynthetic analysis and the investigation of synthetic route were based on the originally proposed structure (yaku’amide A’).

(2) The “PG” represents protecting groups. Ideally, all alcohols would be protected with the same group, and all amines would be protected with another group.


(8) The absolute configurations of those isomers were determined via chemical derivatization to known compounds by J. Jiang and J. M. Cardon.

(10) Normally, MS is not a qualitative method to analyze molecular ratios. However, our observation of this coupling reaction showed that the mass ratio is very good reflection to the real ratio detected in NMR.


(12) For a recent review covering the application of azlactone ring-opening chemistry in peptide synthesis, see: Jiang, J.; Ma, Z.; Castle, S. L. Tetrahedron 2015, 71, 5431–5451.

(13) The carboxylic acid 175 was synthesized using the same chemistry for 157 (Scheme 4.4).

(14) The synthesis of (2S,3R)-OHIle was investigated by J. M. Cardon.

(15) Due to the time limit, characterization of these compounds has not been achieved.
Chapter 5. EXPERIMENTAL SECTION

5.1 General experimental details

Dimethylformamide, methanol, and tetrahydrofuran were dried by passage through a solvent drying system containing cylinders of activated alumina. Flash chromatography was carried out using 60–230 mesh silica gel. $^1$H NMR spectra were acquired on a 500 MHz spectrometer with chloroform (7.27 ppm), methanol (3.34 ppm), or benzene (7.15 ppm) as internal reference. Signals are reported as follows: s (singlet), d (doublet), t (triplet), q (quartet), dd (doublet of doublets), td (triplet of doublets), br s (broad singlet), m (multiplet). Coupling constants are reported in hertz (Hz). $^{13}$C NMR spectra were acquired on a spectrometer operating at 125 MHz with chloroform (77.23 ppm), methanol (49.86 ppm), or benzene (128.62 ppm) as internal reference. Infrared spectra were obtained on an FT-IR spectrometer. Mass spectral data were obtained using ESI techniques.

**General Procedure for Base-Free Aminohydroxylations.** A solution of benzyl 4-chlorobenzoxyloxy carbamate$^2$ (51a, 163.2 mg, 0.534 mmol, 1.7 equiv) in CH$_3$CN (4 mL) at rt was treated with OsO$_4$ (4 wt % solution in H$_2$O, 100 μL, 0.0157 mmol, 0.05 equiv), stirred for 10 min, then treated with the alkene (1 equiv) and H$_2$O (400 μL). A color change from clear to brown typically accompanied addition of the alkene. The resulting mixture was stirred at either rt, 35 °C, or 45 °C for 20–24 h, then treated with sat aq K$_2$S$_2$O$_5$ (400 μL) and stirred for an additional 5 min. It was then diluted with H$_2$O (15 mL) and extracted with EtOAc (3 × 15 mL).
The combined organic layers were washed with sat aq NaHCO$_3$ (2 X 15 mL) and brine (15 mL), dried (MgSO$_4$), and concentrated in vacuo. Flash chromatography (SiO$_2$) afforded the amino alcohol products.

5.2 Experimental procedures and spectral data

Benzyl 1,3-dihydroxy-3-methylbutan-2-ylcarbamate (52a). Prepared from 51a (168.1 mg, 0.550 mmol) and prenol (49, 32 μL, 27.1 mg, 0.315 mmol) according to the General Procedure with stirring at rt for 23.5 h. Flash chromatography (SiO$_2$, 1.5 × 8.5 cm, 5–10% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 52a (49.6 mg, 0.196 mmol, 62%) as a colorless oil. Spectral data were in accord with previously reported data.\(^3\)

**tert-Butyl 1,3-dihydroxy-3-methylbutan-2-ylcarbamate (52b).** Prepared from tert-butyl 4-chlorobenzoyloxy carbamate\(^2\) (147.1 mg, 0.541 mmol) and prenol (49, 32 μL, 27.1 mg, 0.315 mmol) according to the General Procedure with stirring at rt for 24 h. Flash chromatography (SiO$_2$, 1.5 × 8 cm, 5–10% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 52b (51.6 mg, 0.235 mmol, 75%) as a colorless oil. Spectral data were in accord with previously reported data.\(^4\)

Benzyl (2,4-dihydroxy-2-methylbutyl)carbamate (53). Prepared from 51 (167.9 mg, 0.549 mmol) and isoprenol (50, 32 μL, 27.6 mg, 0.321 mmol) according to the General
Procedure with stirring at 35 ºC for 23 h. Flash chromatography (SiO₂, 1.5 × 10 cm, 2–5% MeOH in CH₂Cl₂ gradient elution) afforded 53 (69.1 mg, 0.273 mmol, 85%) as a colorless oil: 

1H NMR (CDCl₃, 500 MHz) δ 7.38–7.28 (m, 5H), 5.47 (s, 1H), 5.09 (s, 2H), 3.94–3.87 (m, 1H), 3.84–3.78 (m, 1H), 3.73 (br s, 1H), 3.24 (dd, J = 13.8, 6.1 Hz, 1H) 3.18 (dd, J = 13.8, 6.3 Hz, 1H), 3.11 (br s, 1H), 1.85–1.77 (m, 1H), 1.63–1.56 (m, 1H), 1.21 (s, 3H); 13C NMR (CDCl₃, 125 MHz) δ 157.5, 136.4, 128.5 (2C), 128.2, 128.1 (2C), 73.3, 66.9, 59.3, 51.0, 39.4, 24.7; IR (film) νmax 3346, 3066, 2935, 1701, 1535, 1455, 1257, 1144, 1042 cm⁻¹; HRMS (ESI) m/z 254.1406 (MH⁺, C₁₃H₁₉NO₄H⁺ requires 254.1387).

**Benzyl (3-hydroxy-1-methoxy-3-methylbutan-2-yl)carbamate (55).** Prepared from 51a (163.5 mg, 0.535 mmol) and 54 (34 μL, 31.6 mg, 0.315 mmol) according to the General Procedure with stirring at 35 ºC for 23.5 h. Flash chromatography (SiO₂, 1.5 × 12 cm, 10–30% EtOAc in hexanes gradient elution) afforded 55 (45.3 mg, 0.169 mmol, 54%) as a colorless oil: 1H NMR (CDCl₃, 500 MHz) δ 7.39–7.29 (m, 5H), 5.62 (d, J = 8.3 Hz, 1H), 5.12 (s, 2H), 3.82 (d, J = 7.2 Hz, 1H), 3.60–3.55 (m, 2H), 3.35 (s, 3H), 3.11 (s, 1H), 1.32 (s, 3H), 1.12 (3H); 13C NMR (CDCl₃, 125 MHz) δ 156.5, 136.4, 128.5 (2C), 128.1, 128.0 (2C), 73.7, 72.8, 66.8, 59.4, 57.0, 27.7, 26.9; IR (film) νmax 3322, 2977, 1701, 1522, 1454, 1216, 1117, 1047 cm⁻¹; HRMS (ESI) m/z 268.1560 (MH⁺, C₁₄H₂₁NO₄H⁺ requires 268.1543).

**Ethyl 2-(((benzyloxy)carbonyl)amino)-3-hydroxy-3-methylbutanoate (57).** Prepared from 51a (492.8 mg, 1.61 mmol) and ethyl 3,3-dimethylacrylate (56, 132 μL, 122 mg, 0.951 mmol) according to the General Procedure with 600 μL OsO₄ solution (0.0944 mmol, 0.10 equiv) and stirring at 35 ºC for 10 h. Flash chromatography (SiO₂, 1.5 × 12 cm, 10–50% EtOAc
in hexanes gradient elution) afforded 57 (221.9 mg, 0.751 mmol, 79%) as a colorless oil: \(^1\)H NMR (CDCl\(_3\), 500 MHz) \(\delta\) 7.39–7.31 (m, 5H), 5.62 (br s, 1H), 5.13 (s, 2H), 4.31–4.19 (m, 3H), 2.50 (br s, 1H), 1.34–1.26 (m, 9H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \(\delta\) 171.6, 156.5, 136.1, 128.5 (2C), 128.2, 128.1 (2C), 71.9, 67.2, 61.62, 61.56, 26.8, 26.3, 14.1; IR (film) \(\nu\)\(_{\text{max}}\) 3406, 2978, 2922, 1720, 1709, 1512, 1501, 1467, 1452, 1211, 1051, 1027 cm\(^{-1}\); HRMS (ESI) \(m/z\) 296.1497 (MH\(^+\), \(\text{C}_{15}\text{H}_{21}\text{NO}_5\text{H}\^+\) requires 296.1492).

**Benzyl (1,4-dihydroxy-4-methylpentan-3-yl)carbamate (59).** Prepared from 51a (168.8 mg, 0.552 mmol) and 58 (37 \(\mu\)L, 31.7 mg, 0.317 mmol) according to the General Procedure with stirring at 35 ºC for 23 h. Flash chromatography (SiO\(_2\), 1.5 \(\times\) 10 cm, 2–5% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 59 (49.5 mg, 0.185 mmol, 58%) as a colorless oil: \(^1\)H NMR (CDCl\(_3\), 500 MHz) \(\delta\) 7.37–7.28 (m, 5H), 5.33 (d, \(J = 9.4\) Hz, 1H), 5.13 (d, \(J = 12.2\) Hz, 1H), 5.08 (d, \(J = 12.2\) Hz, 1H), 3.71–3.64 (m, 2H), 3.63–3.56 (m, 1H), 3.38 (br s, 1H), 2.78 (br s, 1H), 1.98–1.89 (m, 1H), 1.57–1.48 (m, 1H), 1.24 (s, 6H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \(\delta\) 157.7, 136.3, 128.6 (2C), 128.2, 128.0 (2C), 72.2, 67.1, 58.6, 56.1, 32.2, 27.6, 27.0; IR (film) \(\nu\)\(_{\text{max}}\) 3330, 2973, 1697, 1535, 1258, 1054 cm\(^{-1}\); HRMS (ESI) \(m/z\) 268.1561 (MH\(^+\), \(\text{C}_{14}\text{H}_{21}\text{NO}_4\text{H}\^+\) requires 268.1543).

**Benzyl ((2S*,3S*)-1,3-dihydroxy-3-phenylbutan-2-yl)carbamate (61).** Prepared from 51a (116.4 mg, 0.381 mmol) and 60\(^6\) (33.2 mg, 0.224 mmol) according to the General Procedure with stirring at 35 ºC for 18 h. Flash chromatography (SiO\(_2\), 1.5 \(\times\) 11 cm, 2–5% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 17 (32.4 mg, 0.103 mmol, 46%) as a colorless
oil: $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.50–7.42 (m, 3H), 7.41–7.25 (m, 5H), 7.24–7.17 (m, 2H), 5.44 (d, $J = 7.6$ Hz, 1H), 4.98 (s, 2H), 4.18–4.07 (m, 1H), 4.03–3.91 (m, 2H), 3.37 (br s, 1H), 2.28 (br s, 1H), 1.71 (s, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 156.4, 145.0, 136.3, 128.5 (2C), 128.3 (2C), 128.0, 127.7 (2C), 127.1, 124.7 (2C), 77.0 (observed by CDCl$_3$), 66.6, 63.1, 58.9, 28.3; IR (film) $\nu_{\text{max}}$ 3404, 2977, 1701, 1517, 1447, 1251, 1072, 1048 cm$^{-1}$; HRMS (ESI) $m/z$ 316.1558 (MH$^+$, C$_{18}$H$_{21}$NO$_4$H$^+$ requires 316.1543.

Benzyl ((2$S$*,3$S$*)-1,3-dihydroxy-3-methylpentan-2-yl)carbamate (63).

Prepared from 51a (164.2 mg, 0.537 mmol) and 62 (37.5 μL, 27.2 mg, 0.316 mmol) according to the General Procedure with 0.0315 mmol, 0.10 equiv) and stirring at 35 ºC for 23 h. Flash chromatography (SiO$_2$, 1.5 × 11 cm, 0.1–2% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 63 (46.6 mg, 0.174 mmol, 55%) as a colorless oil: $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.38–7.30 (m, 5H), 5.63 (d, $J = 8.1$ Hz, 1H), 5.12 (s, 2H), 4.06–4.00 (m, 1H), 3.88–3.82 (m, 1H), 3.62–3.57 (m, 1H), 2.63 (br s, 1H), 2.54 (br s, 1H), 1.63–1.55 (m, 1H), 1.54–1.45 (m, 1H), 1.29 (s, 3H), 0.89 (t, $J = 7.5$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 156.7, 136.4, 128.1, 128.0 (2C), 75.8, 66.9, 63.6, 56.0, 32.3, 32.3, 23.9, 8.1; IR (film) $\nu_{\text{max}}$ 3400, 2970, 1701, 1517, 1447, 1251, 1072, 1048 cm$^{-1}$; HRMS (ESI) $m/z$ 268.1557 (MH$^+$, C$_{14}$H$_{21}$NO$_4$H$^+$ requires 268.1543.

Benzyl ((2$S$*,3$R$*)-1,3-dihydroxy-3-methylpentan-2-yl)carbamate (65).

Prepared from 51a (163.7 mg, 0.536 mmol) and 64 (37 μL, 31.3 mg, 0.312 mmol) according to the General Procedure with 0.0315 mmol, 0.10 equiv) and stirring at 35 ºC for 23 h. Flash chromatography (SiO$_2$, 1.5 × 11 cm, 0.1–2% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 65 (48.9 mg, 0.183 mmol, 59%) as a colorless oil: $^1$H NMR (CDCl$_3$, 500 MHz)
7.40–7.30 (m, 5H), 5.69 (br s, 1H), 5.11 (s, 2H), 4.04–3.98 (m, 1H), 3.83–3.77 (m, 1H), 3.61–3.57 (m, 1H), 2.77–2.55 (m, 2H), 1.77–1.58 (m, 2H), 1.16 (s, 3H), 0.95 (t, \( J = 7.5 \) Hz, 3H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \(\delta\) 156.7, 136.4, 128.5 (2C), 128.2 (2C), 128.0, 76.1, 66.9, 63.1, 56.6, 32.8, 23.4, 8.3; IR (film) \(\nu_{\max}\) 3401, 2971, 1701, 1522, 1455, 1251, 1062 cm\(^{-1}\); HRMS (ESI) \(m/z\) 268.1543 (MH\(^+\), C\(_{14}\)H\(_{21}\)NO\(_4\)H\(^+\) requires 268.1543).

**Benzyl 2,3-dihydroxy-2-methylpropylcarbamate (67).** Prepared from 51a (163.9 mg, 0.536 mmol) and allylic alcohol 66 (27 \(\mu\)L, 23.0 mg, 0.319 mmol) according to the General Procedure with stirring at rt for 23 h. Flash chromatography (SiO\(_2\), 1.5 \(\times\) 10 cm, 5–10% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 67 (47.7 mg, 0.199 mmol, 62%) as a colorless oil. Spectral data were in accord with previously reported data.\(^9\)

**Benzyl 2,3-dihydroxypropylcarbamate (69).** Prepared from 51a (164.5 mg, 0.538 mmol) and allyl alcohol (68, 21.5 \(\mu\)L, 18.4 mg, 0.316 mmol) according to the General Procedure with stirring at rt for 23.5 h. Flash chromatography (SiO\(_2\), 1.5 \(\times\) 9.5 cm, 5–10% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 69 (49.6 mg, 0.220 mmol, 70%) as a colorless oil that was a 6.7:1 ratio of regioisomers. Spectral data for both 69 and its meso regioisomer were in accord with previously reported data.\(^10\)

**Ethyl 2-amino-3-hydroxy-3-methylbutanoate (57a).** A suspension of 57 (280.4 mg, 0.949 mmol) and Pd/C (10 wt %, 42.6 mg) in MeOH (12 mL) was stirred at rt under H\(_2\) (450 psi) for 23 h. The mixture was filtered through Celite, and the Celite pad was washed with MeOH
(125 mL). The filtrate was concentrated in vacuo to afford 57a as a colorless oil, which was used directly in the next step without further purification: \(^1\)H NMR (CDCl\(_3\), 500 MHz) \(\delta\) 4.29–4.16 (m, 2H), 3.14 (s, 1H), 2.75 (br s, 3H), 1.32 (s, 3H), 1.30 (t, \(J = 7.1\) Hz, 3H), 1.05 (s, 3H); HRMS (ESI) \(m/z\) 162.1105 (MH\(^+\), C\(_7\)H\(_{15}\)NO\(_3\)H\(^+\) requires 162.1125).

**Ethyl 2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-hydroxy-3-methylbutanoate (70).** A solution of above amine 57a (0.949 mmol) in THF–DMF (2.5:1, 42.5 mL) at 0 °C under Ar was treated with N-Cbz-glycine (370.0 mg, 1.769 mmol), HOBt (298.9 mg, 1.770 mmol), and EDC•HCl (338.5 mg, 1.766 mmol). The resulting mixture was allowed to warm to rt and stir for 24 h. The reaction was quenched with sat aq NaHCO\(_3\) (15 mL), and the resulting precipitate was filtered and washed with EtOAc (15 mL). The aqueous layer was extracted with EtOAc (3 × 15 mL), and the combined organic extracts were washed with brine (15 mL), dried (Na\(_2\)SO\(_4\)), filtered, and concentrated in vacuo. Flash chromatography (SiO\(_2\), 2.3 × 17 cm, 1–7% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 70 (286.4 mg, 0.812 mmol, 86% over 2 steps) as a colorless oil: \(^1\)H NMR (CDCl\(_3\), 500 MHz) \(\delta\) 7.40–7.30 (m, 5H), 6.86 (d, \(J = 8.2\) Hz, 1H), 5.48 (br s, 1H), 5.14 (s, 2H), 4.52 (d, \(J = 8.8\) Hz, 1H), 4.30–4.15 (m, 2H), 3.94 (d, \(J = 5.4\) Hz, 2H), 2.71 (br s, 1H), 1.30 (t, \(J = 7.2\) Hz, 3H), 1.28 (s, 3H), 1.24 (s, 3H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \(\delta\) 171.2, 169.2, 156.6, 136.1, 128.6 (2C), 128.3, 128.1 (2C), 71.9, 67.3, 61.7, 59.8, 44.5, 26.7, 26.6, 14.1; IR (film) \(\nu_{\text{max}}\) 3342, 2980, 1731, 1531, 1455, 1374, 1261, 1028 cm\(^{-1}\); HRMS (ESI) \(m/z\) 353.1716 (MH\(^+\), C\(_{17}\)H\(_{24}\)N\(_2\)O\(_6\)H\(^+\) requires 353.1707).
Ethyl 2-(2-(benzyloxycarbonylamino)acetamido)-3-methylbut-2-enoate (71a). A solution of alcohol 70 (20.9 mg, 0.059 mmol) in chloroform (250 μL) was treated with a solution of Martin sulfurane (500 μL, 0.117 mmol, 0.23 M in CHCl₃). The resultant mixture was heated to 50 ºC and stirred for 1 h. The reaction mixture was cooled to rt and concentrated in vacuo. Flash chromatography (SiO₂, 1.5 × 7 cm, 0–1.6% MeOH in CH₂Cl₂ gradient elution) afforded 71a (15.9 mg, 0.048 mmol, 80%) as a colorless oil: ¹H NMR (13.4:1 mixture of rotomers, data for major rotomer, CDCl₃, 500 MHz) δ 7.38–7.32 (m, 5H), 7.15 (br s, 1H), 5.42 (br s, 1H), 5.15 (s, 2H), 4.20 (q, J = 7.1 Hz, 2H), 3.96 (d, J = 5.7 Hz, 2H), 2.18 (s, 3H), 1.82 (s, 3H), 1.27 (t, J = 7.1 Hz, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 167.7, 164.6, 156.7, 146.5, 136.0, 128.6 (2C), 128.3, 128.1 (2C), 120.5, 67.3, 60.9, 44.7, 22.7, 21.4, 14.1; IR (film) ν_max 3316, 2923, 2853, 1714, 1514, 1457, 1309, 1237, 1093; HRMS (ESI) m/z 335.1592 (MH⁺, C₁₇H₂₂N₂O₅H⁺ requires 335.1601).

Ethyl (2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-hydroxy-3-methylbutanoyl)glycinate (73). A solution of ester 69 (105 mg, 0.298 mmol) in MeOH–H₂O (1:1, 2 mL) at 0 ºC was treated with LiOH•H₂O (90 mg, 2.14 mmol, 7.2 equiv), then stirred at 0 ºC for 3 h. The resulting mixture was acidified to pH 3 by the addition of 10% citric acid (5 mL) and dissolved in EtOAc (60 mL), then washed with H₂O (30 mL × 2) and brine (30 mL × 2). The combined organic layers were dried (Na₂SO₄) and concentrated in vacuo. The crude carboxylic acid (68 mg, 0.210 mmol, 70%) was used directly without further purification.
The acid was dissolved in CH2Cl2–DMF (2.8 mL–0.2mL) was treated with Ethyl glycine HCl salt (59.9 mg, 0.429 mmol, 2.0 equiv), HOBt (ca. 14% H2O content, 58.4 mg, 0.327 mmol, 1.6 equiv), and EDC•HCl (57.4 mg, 0.299 mmol, 1.4 equiv), Na2CO3 (24.3 mg, 0.229 mmol, 1.1 equiv) at 0 °C under Ar. The resulting mixture was stirred at 0 °C to rt under Ar for 11 h. The reaction was treated with H2O (6 mL), then extracted with EtOAc (5 × 6 mL), and the combined organic layers were washed with brine (10 mL), dried (Na2SO4), and concentrated in vacuo.

Flash chromatography (7 mL of SiO2, 1–4% MeOH in CH2Cl2 gradient elution) afforded 80 (45 mg, 0.109 mmol, 52%, 36% for two steps) as a colorless oil: 1H NMR (CDCl3, 500 MHz) of major rotamer: δ 7.42–7.32 (m, 5H), 7.23 (br s, 1H), 7.13 (d, J = 8.4 Hz, 1H), 5. 7 (s, 1H), 5.14 (s, 2H), 4.42 (d, J = 8.7 Hz, 1H), 4.21 (q, J = 7.29 Hz, 2H), 4.08–3.87 (m, 2H), 1.35 (s, 3H), 1.29 (t, J = 7.2 Hz, 3H), 1.20 (s, 3H); 13C NMR (CDCl3, 125 MHz) δ 171.5, 169.8, 169.6, 156.7, 136.1, 128.6 (2C), 128.3, 128.2 (2C), 71.8, 67.3, 61.7, 59.0, 44.5, 41.2, 27.3, 25.5, 14.1; HRMS (ESI) m/z 410.2161 (MH+, C18H28N3O7H+ requires 410.1927).

Ethyl (2S*,3R*)-2-(((Benzyloxy)carbonyl)amino)-3-hydroxy-3-methylpentanoate (76). A solution of benzyl ((methylsulfonyl)oxy)carbamate11 (606.1 mg, 2.471 mmol, 1.5 equiv) in CH3CN (18 mL) at rt was treated with OsO4 (4 wt % solution in H2O, 1.0 mL, 0.16 mmol, 0.10 equiv), stirred for 10 min, and then treated with a solution of enoate 7512 (234.9 mg, 1.652 mmol) in CH3CN (9 mL) and H2O (2.4 mL). The resulting mixture was stirred at 35 °C for 4 d, treated with sat aq K2S2O5 (3.6 mL), and stirred for an additional 5 min. It was then diluted with H2O (15 mL) and extracted with EtOAc (6 × 15 mL). The combined organic layers were washed with sat aq NaHCO3 (2 × 50 mL) and brine (15 mL), dried (Na2SO4), and concentrated in vacuo. Flash chromatography (ca. 80 mL of SiO2, 0.5–1 % MeOH in
CH$_2$Cl$_2$ gradient elution) afforded a mixture of 76 (356.3 mg, 1.152 mmol, 70%) and benzyl carbamate (170.8 mg) as a light yellow oil. This mixture could be used directly in the subsequent hydrogenolysis reaction with no complications. An analytical sample of 76 could be obtained after further chromatography. For 76: $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.35–7.29 (m, 5H), 5.75 (d, $J$ = 8.5 Hz, 1H), 5.10 (s, 2H), 4.30 (d, $J$ = 9.0 Hz, 1H), 4.27–4.17 (m, 2H), 2.58 (s, 1H), 1.56 (qd, $J$ = 7.5, 1.8 Hz, 2H), 1.29 (t, $J$ = 6.9 Hz, 3H), 1.19 (s, 3H), 0.92 (t, $J$ = 7.2 Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 171.9, 156.4, 136.1, 128.5 (2C), 128.2 (2C), 128.1, 73.9, 67.2, 61.5, 59.9, 31.3, 23.4, 14.1, 7.9; IR (film) $\nu_{\max}$ 3434, 2977, 1724, 1516, 1206, 1061 cm$^{-1}$; HRMS (ESI) $m/z$ 310.1632 (MH$^+$, C$_{16}$H$_{23}$NO$_3$H$^+$ requires 310.1654).

**Ethyl (2S*,3R*)-2-(2-(((Benzyloxy)carbonyl)amino)acetamido)-3-hydroxy-3-methyloctanoate (77).** A solution of 76 (356.3 mg contaminated with 170.8 mg of benzyl carbamate, 1.152 mmol) in MeOH (15 mL) was treated with 10% Pd/C (100 mg, 0.28 wt equiv) and stirred at rt under H$_2$ (500 psi) for 3 d.$^{13}$ The mixture was filtered through a pad of Celite (washed with 125 mL of MeOH), and the filtrate was concentrated in vacuo to afford the crude amine, which was used without further purification.

A solution of the amine in anhydrous THF–DMF (3:1, 24 mL) at 0 °C under Ar was treated with N-Cbz-glycine (473.4 mg, 2.263 mmol), HOBt (ca. 14% H$_2$O content, 390.1 mg, 2.483 mmol), and EDC•HCl (430.2 mg, 2.244 mmol). The resulting mixture stirred at 0 °C to rt under Ar for 24 h. The reaction was quenched by the addition of sat aq NaHCO$_3$ (20 mL) and diluted with H$_2$O (20 mL). The aqueous layer was extracted with EtOAc (4 × 40 mL), and the combined organic extracts were washed with brine (40 mL), dried (Na$_2$SO$_4$), and concentrated in
Flash chromatography (90 mL of SiO$_2$, 0–3% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 77 (332.3 mg, 0.9069 mmol, 79%) as a colorless oil: $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.40–7.29 (m, 5H), 6.81 (d, $J$ = 8.4 Hz, 1H), 5.47 (br s, 1H), 5.14 (s, 2H), 4.56 (d, $J$ = 8.8 Hz, 1H), 4.27–4.16 (m, 2H), 3.99–3.88 (m, 2H), 2.60 (br s, 1H), 1.53 (q, $J$ = 7.0 Hz, 2H), 1.30 (t, $J$ = 7.1 Hz, 3H), 1.20 (s, 3H), 0.92 (t, $J$ = 6.7 Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 171.5, 169.1, 156.5, 136.1, 128.5 (2C), 128.2 (2C), 128.1, 74.1, 67.2, 61.7, 58.0, 44.4, 31.6, 23.4, 14.1, 7.9; IR (film) $\nu_{\text{max}}$ 3346, 2977, 1731, 1526, 1261 cm$^{-1}$; HRMS (ESI) $m/z$ 367.1868 (MH$^+$, C$_{18}$H$_{26}$N$_2$O$_6$H$^+$ requires 367.1869).

Ethyl (2$S$*,3$S$*)-2-(((Benzyloxy)carbonyl)amino)-3-hydroxy-3-methylpentanoate (79). A solution of benzyl ((methylsulfonyl)oxy)carbamate$^2$ (820 mg, 3.34 mmol, 1.4 equiv) in CH$_3$CN (18 mL) at rt was treated with OsO$_4$ (4 wt % solution in H$_2$O, 1.5 mL, 0.24 mmol, 0.099 equiv), stirred for 10 min, and then treated with a solution of enoate 78$^{14}$ (340 mg, 2.39 mmol) in CH$_3$CN (6.0 mL) and H$_2$O (1.5 mL). The resulting mixture was stirred at 45 °C for 2.5 d, treated with sat aq K$_2$S$_2$O$_5$ (7 mL), and stirred for an additional 10 min. It was then diluted with H$_2$O (35 mL) and extracted with EtOAc (3 × 60 mL). The combined organic layers were washed with sat aq NaHCO$_3$ (2 × 50 mL) and brine (40 mL), dried (Na$_2$SO$_4$), and concentrated in vacuo. Flash chromatography (100 mL of SiO$_2$, 1–5% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 79 (580 mg, 1.87 mmol, 78%) as a light yellow oil: $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.37–7.30 (m, 5H), 5.62 (d, $J$ = 9.3 Hz, 1H), 5.12 (s, 2H), 4.31 (d, $J$ = 9.4 Hz, 1H), 4.28–4.18 (m, 2H), 2.39 (br s, 1H), 1.56–1.46 (m, 2H), 1.30 (t, $J$ = 7.2 Hz, 3H), 1.18 (s, 3H), 0.98 (t, $J$ = 7.2 Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 171.9, 156.3, 136.1, 128.5 (2C), 128.2
(2C), 128.1, 74.3, 67.2, 61.6, 60.3, 32.5, 22.3, 14.1, 8.0; IR (film) $\nu_{\text{max}}$ 3363, 2978, 1716, 1519, 1337, 1208, 1058 cm$^{-1}$; HRMS (ESI) $m/z$ 310.1711 (MH$^+$, C$_{16}$H$_{23}$NO$_5$H$^+$ requires 310.1654).

**Ethyl (25*,35*)-2-(2-(((Benzyloxy)carbonyl)amino)acetamido)-3-hydroxy-3-methylpentanoate (80).** A solution of 79 (353 mg, 1.14 mmol) in MeOH (10 mL) was treated with 10% Pd/C (114.6 mg, 0.32 wt equiv) and stirred at rt under H$_2$ (550 psi) for 2.5 d. The mixture was filtered through a pad of Celite (washed with 125 mL of MeOH), and the filtrate was concentrated *in vacuo* to afford the crude amine (198 mg, 1.13 mmol), a portion of which was used without further purification.

A solution of the amine (170 mg, 0.970 mmol) in anhydrous DMF (12 mL) at 0 °C under Ar was treated with N-Cbz-glycine (401.6 mg, 1.92 mmol, 2.0 equiv), HOBt (ca. 14% H$_2$O content, 327.5 mg, 2.08 mmol, 2.1 equiv), and EDC•HCl (375.7 mg, 1.96 mmol, 2.0 equiv). The resulting mixture was stirred at 0 °C to rt under Ar for 48 h. The reaction was quenched by the addition of sat aq NaHCO$_3$ (12 mL) and diluted with H$_2$O (12 mL). The aqueous layer was extracted with EtOAc (3 × 30 mL), and the combined organic layers were washed with brine (20 mL), dried (Na$_2$SO$_4$), and concentrated *in vacuo*. Flash chromatography (70 mL of SiO$_2$, 1–5% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 80 (220 mg, 0.600 mmol, 62%) as a colorless oil: $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.38–7.30 (m, 5H), 6.74 (d, $J = 8.4$ Hz, 1H), 5.37 (s, 1H), 5.14 (s, 2H), 4.56 (d, $J = 8.9$ Hz, 1H), 4.28–4.16 (m, 2H), 3.98–3.89 (m, 2H), 2.42 (br s, 1H), 1.51 (q, $J = 7.1$ Hz, 2H), 1.30 (t, $J = 7.2$ Hz, 3H), 1.14 (s, 3H), 0.98 (t, $J = 7.5$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 171.4, 169.0, 156.5, 136.0, 128.5 (2C), 128.3, 128.2 (2C), 74.3, 67.3, 61.7, 58.4, 44.5, 32.5, 22.4, 14.1, 8.0; IR (film) $\nu_{\text{max}}$ 3344, 2979, 1728, 1525, 1212, 1051 cm$^{-1}$; HRMS (ESI) $m/z$ 367.1862 (MH$^+$, C$_{18}$H$_{26}$N$_2$O$_6$H$^+$ requires 367.1869).
Ethyl (Z)-2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-methylpent-2-enoate (82). A solution of alcohol 77 (19.5 mg, 0.0532 mmol) in anhydrous CHCl₃ (180 μL) was treated with Martin sulfurane (0.21 M in anhydrous CHCl₃, 500 μL, 0.105 mmol, 2.0 equiv). The resulting mixture was stirred at 50 °C under Ar for 1 h, cooled to rt, and concentrated in vacuo. Flash chromatography (5 mL of SiO₂, 0–1.6% MeOH in CH₂Cl₂ gradient elution) afforded 82 (16.2 mg, 0.0465 mmol, 87%, >19:1 dr) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz, 12:1 mixture of rotamers, data for major rotamer) δ 7.40–7.30 (m, 5H), 7.08 (br s, 1H), 5.38 (br s, 1H), 5.15 (s, 2H), 4.19 (q, J = 7.1 Hz, 2H), 3.95 (d, J = 5.7 Hz, 2H), 2.20–2.09 (m, 5H), 1.27 (t, J = 7.2 Hz, 3H), 1.02 (t, J = 7.3 Hz, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 168.2, 164.7, 156.7, 151.0, 136.0, 128.6 (2C), 128.3, 128.1 (2C), 120.1, 67.3, 60.9, 44.8, 28.7, 18.6, 14.2, 11.5; IR (film) νₘₐₓ 3313, 2926, 1718, 1509, 1216 cm⁻¹; HRMS (ESI) m/z 349.1777 (MH⁺, C₁₈H₂₄N₂O₅H⁺ requires 349.1763).

Ethyl (E)-2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-methylpent-2-enoate (105). A solution of alcohol 80 (34.6 mg, 0.0.0944 mmol) in anhydrous CHCl₃ (0.5 mL) was treated with Martin sulfurane (0.41 M in anhydrous CHCl₃, 0.5 mL, 0.21 mmol, 2.2 equiv). The resulting mixture was stirred at 50 °C under Ar for 1 h, cooled to rt, and concentrated in vacuo. Flash chromatography (8 mL of SiO₂, 0–2.6% MeOH in CH₂Cl₂ gradient elution) afforded 105 (24.1 mg, 0.0692 mmol, 73%, >19:1 dr) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz, 15:1 mixture of rotamers, data for major rotamer) δ 7.40–7.30 (m, 5H), 7.13 (s, 1H), 5.39 (s, 1H), 5.15 (s, 2H), 4.20 (q, J = 7.1 Hz, 2H), 3.95 (d, J = 5.6 Hz, 2H), 2.53 (q, J = 7.5 Hz, 2H),
1.79 (s, 3H), 1.27 (t, \( J = 7.1 \) Hz, 3H), 1.10 (t, \( J = 7.5 \) Hz, 3H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \( \delta \) 167.4, 164.3, 156.7, 150.6, 136.0, 128.6 (2C), 128.3, 128.1 (2C), 120.3, 67.4, 60.9, 44.8, 27.7, 19.9, 14.2, 12.7; IR (film) \( \nu_{\text{max}} \) 3311, 2977, 2934, 1720, 1514, 1266, 1211 cm\(^{-1}\); HRMS (ESI) \( m/z \) 349.1858 (MH\(^+\), C\(_{18}\)H\(_{24}\)N\(_2\)O\(_5\)H\(^+\) requires 349.1763).

Benzyl (E)-(2-((1-((2-hydroxyethyl)amino)-3-methyl-1-oxo-pent-2-en-2-yl)amino)-2-oxo-ethyl)carbamate (110). A solution of azide 122a (3.0 mg, 0.0077 mmol) in THF (210 \( \mu \)L) and H\(_2\)O (16 \( \mu \)L) at 0 \( ^\circ \)C under Ar was treated dropwise with PMe\(_3\) (1 M in THF, 23 \( \mu \)L, 0.023 mmol, 3.0 equiv). The resulting mixture was stirred at 0 \( ^\circ \)C to rt for 20 h, at which time the starting material had disappeared as evidenced by MS. The mixture was then treated dropwise with morpholine (24 \( \mu \)L, 24 mg, 0.28 mmol), and stirred at rt for 60 h followed by concentration \textit{in vacuo}. The residue was dissolved in EtOAc (5 mL), washed with H\(_2\)O (2 \( \times \) 1 mL) and brine (1 mL), dried (Na\(_2\)SO\(_4\)), and concentrated \textit{in vacuo}. Flash chromatography (2 mL of SiO\(_2\), 0–3% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 110 (2.7 mg, 0.0074 mmol, 96%, 10:1 dr) as a white film: \(^1\)H NMR (CDCl\(_3\), 500 MHz) \( \delta \) 7.40 (br s, 1H), 7.39–7.31 (m, 5H), 6.59 (br s, 1H), 5.46 (br s, 1H), 5.14 (s, 2H), 3.89 (d, \( J = 5.7 \) Hz, 2H), 3.73 (q, \( J = 5.1 \) Hz, 2H), 3.43 (q, \( J = 5.0 \) Hz, 2H), 3.24 (br s, 1H), 2.41 (q, \( J = 7.3 \) Hz, 2H), 1.70 (s, 3H), 1.09 (t, \( J = 7.5 \) Hz, 3H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \( \delta \) 168.4, 166.6, 157.2, 142.4, 135.6, 128.6 (2C), 128.5, 128.2 (2C), 123.7, 67.7, 61.4, 45.1, 42.7, 27.0, 17.5, 12.8; IR (film) \( \nu_{\text{max}} \) 3316, 2919, 1685, 1522, 1248, 1050 cm\(^{-1}\); HRMS (ESI) \( m/z \) 364.1871 (MH\(^+\), C\(_{18}\)H\(_{25}\)N\(_3\)O\(_5\)H\(^+\) requires 364.1872).
A solution of ester 15 (541 mg, 1.48 mmol) in t-BuOH–H₂O (3:1, 5.3 mL) at 0 °C was treated with LiOH•H₂O (310 mg, 7.39 mmol, 5.0 equiv), then stirred at 0 °C for 2 h. The resulting mixture was acidified to pH 1~2 by the addition of 2 N HCl (4 mL) and extracted with EtOAc (6 × 5 mL). The combined organic layers were dried (Na₂SO₄) and concentrated in vacuo. The crude carboxylic acid (490.1 mg, 1.448 mmol, 98%) was used directly without further purification.

A solution of the crude carboxylic acid (36.4 mg, 0.108 mmol) and iodide 22a¹⁵ (45.3 mg, 0.230 mmol, 2.1 equiv) in anhydrous DMF (1 mL) at rt under Ar was treated with Et₃N (46 µL, 33 mg, 0.33 mmol, 3.1 equiv). The resulting mixture was stirred at 80 °C under Ar for 22 h, then concentrated in vacuo. The residue was dissolved in EtOAc (20 mL) and washed with brine (3 × 3 mL). The organic layer was dried (Na₂SO₄) and concentrated in vacuo. Flash chromatography (5 mL of SiO₂, 0–2% MeOH in CH₂Cl₂ gradient elution) afforded 114 (37.5 mg, 0.0920 mmol, 86%, 84% from 77) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz) δ 7.40–7.30 (m, 5H), 6.73 (d, J = 8.1 Hz, 1H), 5.37 (br s, 1H), 5.14 (s, 2H), 4.58 (d, J = 8.6 Hz, 1H), 4.36–4.26 (m, 2H), 3.96–3.90 (m, 2H), 3.61–3.46 (m, 2H), 2.29 (s, 1H), 1.57–1.49 (m, 2H), 1.25 (s, 3H), 0.92 (t, J = 7.1 Hz, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 170.9, 169.1, 156.5, 136.1, 128.6 (2C), 128.3 (2C), 128.1, 73.9, 67.3, 63.7, 58.2, 49.5, 44.5, 31.6, 23.5, 7.9; IR (film) ν_max 3367, 2971, 2106, 1739, 1525, 1270 cm⁻¹; HRMS (ESI) m/z 408.1918 (MH⁺, C₁₈H₂₅N₅O₆H⁺ requires 408.1883).
2-Azidoethyl (Z)-2-((benzyloxy)carbonyl)amino)acetamido)-3-methylpent-2-enolate (115). A solution of alcohol 114 (23.8 mg, 0.0584 mmol) in anhydrous CHCl₃ (250 μL) was treated with Martin sulfurane (0.23 M in anhydrous CHCl₃, 500 μL, 0.12 mmol, 2.0 equiv) dropwise at 0 °C. The resulting mixture was stirred at 0 °C under Ar for 1 h, warmed to rt, and concentrated in vacuo. Flash chromatography (8 mL of SiO₂, 0–1.5% MeOH in CH₂Cl₂ gradient elution) afforded 115 (19.4 mg, 0.0498 mmol, 85%, >19:1 dr) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz) δ 7.40–7.30 (m, 5H), 7.16 (br s, 1H), 5.40 (br s, 1H), 5.15 (s, 2H), 4.29 (t, J = 5.0 Hz, 2H), 3.95 (d, J = 5.9 Hz, 2H), 3.47 (t, J = 4.5 Hz, 2H), 2.22–2.09 (m, 5H), 1.03 (t, J = 6.9 Hz, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 168.4, 163.9, 156.8, 153.0, 136.0, 128.6 (2C), 128.4, 128.2 (2C), 119.5, 67.4, 63.4, 49.8, 44.8, 28.7, 18.7, 11.4; IR (film) νₘₐₓ 3316, 2938, 2108, 1719, 1509, 1259 cm⁻¹; HRMS (ESI) m/z 390.1674 (MH⁺, C₁₈H₂₃N₅O₅H⁺ requires 390.1777).

Benzyl (Z)-(2-((1-((2-hydroxyethyl)amino)-3-methyl-1-oxopent-2-en-2-yl)amino)-2-oxoethyl)carbamate (117). A solution of azide 115 (9.4 mg, 0.0241 mmol) in THF (650 μL) and H₂O (50 μL) at 0 °C under Ar was treated dropwise with PMe₃ (1 M in THF, 72 μL, 0.072 mmol, 3.0 equiv). The resulting mixture was stirred at 0 °C to rt for 21 h, at which time the starting material had disappeared as evidenced by MS. The mixture was then cooled to 0 °C, treated dropwise with morpholine (98 μL, 99 mg, 1.1 mmol), and stirred at 0 °C to rt for 72 h followed by concentration in vacuo. The residue was dissolved in EtOAc (10 mL), washed with H₂O (2 × 2 mL) and brine (2 mL), dried (Na₂SO₄), and concentrated in vacuo. Flash
chromatography (2 mL of SiO$_2$, 0–3% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 117 (7.9 mg, 0.0217 mmol, 90%, 16:1 dr) as a white film: $^1$H NMR (CDCl$_3$, 500 MHz, 20:1 mixture of rotamers, data for major rotamer) $\delta$ 7.74 (br s, 1H), 7.40–7.30 (m, 5H), 6.64 (br s, 1H), 5.48 (br s, 1H), 5.14 (s, 2H), 3.89 (d, $J = 5.6$ Hz, 2H), 3.73 (q, $J = 4.8$ Hz, 2H), 3.44 (q, $J = 5.1$ Hz, 2H), 3.22 (br s, 1H), 2.06 (q, $J = 7.1$ Hz, 2H), 2.02 (s, 3H), 1.01 (t, $J = 7.5$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 169.1, 167.2, 157.2, 142.7, 135.8, 128.6 (2C), 128.4, 128.2 (2C), 123.3, 67.6, 61.4, 44.9, 42.6, 27.0, 17.9, 11.5; IR (film) $\nu$$_{\text{max}}$ 3286, 2925, 1655, 1526, 1236, 1050 cm$^{-1}$; HRMS (ESI) $m/z$ 364.1923 (MH$^+$, C$_{18}$H$_{25}$N$_3$O$_5$H$^+$ requires 364.1872).

(R)-2-azido-1-iodo-3-methylbutane (78b). A solution of (R)-(−)-2-amino-3-methyl-1-butanol (321 µL, 299 mg, 2.88 mmol), K$_2$CO$_3$ (398 mg, 2.88 mmol), and CuSO$_4$•5H$_2$O (7.8 mg, 0.031 mmol) in H$_2$O (9.3 mL) and MeOH (18.6 mL) at rt under Ar was treated with a solution of TfN$_3$ in CH$_2$Cl$_2$ (prepared according to the procedure of Lundquist and Pelletier, ca. 0.42 M, 13.7 mL, ca. 5.8 mmol). The resulting mixture was stirred at rt for 48 h. The layers were separated, and the aqueous layer was extracted with CH$_2$Cl$_2$ (4 × 20 mL). The combined organic layers were washed with brine (20 mL), dried (Na$_2$SO$_4$), and concentrated in vacuo. The crude azido alcohol was used in the next step without further purification.

A solution of PPh$_3$ (937 mg, 3.57 mmol) in anhydrous CH$_2$Cl$_2$ (30 mL) at rt under Ar was treated with imidazole (525 mg, 7.71 mmol) followed by I$_2$ (1.44 g, 5.67 mmol), stirred for 5 min, then treated dropwise with the crude azido alcohol. The resulting mixture was refluxed for 48 h, cooled to rt, and treated with sat aq Na$_2$SO$_3$ (20 mL). It was stirred until the color changed from black to yellow, at which time the layers were separated. The aqueous layer was extracted with CH$_2$Cl$_2$ (4 × 20 mL), and the combined organic layers were washed with brine (20 mL), dried
(Na$_2$SO$_4$), and concentrated in vacuo. Flash chromatography (100 mL of SiO$_2$, 0–0.5% EtOAc in hexanes gradient elution) afforded 78b (459 mg, 1.92 mmol, 67% over 2 steps) as a colorless oil: [α]$^\text{D}_\text{25}$ −5.0 (c 0.62, CHCl$_3$); $^1$H NMR (CDCl$_3$, 500 MHz) δ 3.43–3.24 (m, 3H), 2.08–1.92 (m, 1H), 1.02 (d, $J = 6.9$ Hz, 3H), 1.00 (d, $J = 6.8$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) δ 69.4, 32.9, 19.5, 17.3, 6.9; IR (film) $v_{\text{max}}$ 2965, 2110, 1270 cm$^{-1}$.

(R)-2-azido-3-methylbutyl (2S*,3R*)-2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-hydroxy-3-methylpentanoate (118). A solution of the acid derived from hydrolysis of ester 77 (prepared as described for azidoethyl ester 114a, 33.5 mg, 0.0990 mmol) and iodide 78b (48 mg, 0.20 mmol, 2.0 equiv) in anhydrous DMF (950 μL) at rt under Ar was treated with Et$_3$N (40 μL, 29 mg, 0.29 mmol, 2.9 equiv). The resulting mixture was stirred at 75 °C under Ar for 24 h, then concentrated in vacuo. The residue was dissolved in EtOAc (20 mL) and washed with brine (3 × 3 mL). The organic layer was dried (Na$_2$SO$_4$) and concentrated in vacuo. Flash chromatography (5 mL of SiO$_2$, 0–2% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 118 (32.9 mg, 0.0732 mmol, 74%, 72% from 15) as a yellow oil that was a 1:1 mixture of diastereomers: $^1$H NMR (CDCl$_3$, 500 MHz) δ 7.40–7.29 (m, 5H), 6.82 and 6.80 (2d, $J = 9.5$ Hz, 1H), 5.44 (s, 1H), 5.13 (s, 2H), 4.60 and 4.58 (2d, $J = 8.8$ Hz, 1H), 4.40–4.28 (m, 1H), 4.20–4.08 (m, 1H), 3.97–3.85 (m, 2H), 3.50–3.44 and 3.44–3.36 (2m, 1H), 2.60–2.17 (br s, 1H), 1.92–1.78 (m, 1H), 1.61–1.44 (m, 2H), 1.24 (d, $J = 6.5$ Hz, 3H), 1.02–0.95 (m, 6H), 0.92 (t, $J = 6.4$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) δ 171.1 and 170.8, 169.1, 156.5, 136.1, 128.6 (2C), 128.3 (2C), 128.1, 74.0, 67.3, 66.6, 66.2, 58.4 and 58.3, 44.5, 31.7, 30.1 and 29.9, 23.5 and 23.3, 19.4, 18.2 and 18.1, 8.0 and 7.9; IR (film) $v_{\text{max}}$ 3341, 2969, 2101, 1733, 1522, 1270 cm$^{-1}$; HRMS (ESI) $m/z$ 450.2359 (MH$^+$, C$_{21}$H$_{31}$N$_5$O$_6$H$^+$ requires 450.2353).
(R)-2-azido-3-methylbutyl (Z)-2-((benzylloxy)carbonyl)amino)acetamido)-3-methylpent-2-enoate (119). A solution of alcohol 118 (23.5 mg, 0.0523 mmol) in anhydrous CHCl₃ (220 μL) was treated with Martin sulfurane (0.24 M in anhydrous CHCl₃, 440 μL, 0.11 mmol, 2.0 equiv) dropwise at −20 °C. The resulting mixture was stirred at −20 °C under Ar for 1 h, warmed to rt, and concentrated in vacuo. Flash chromatography (8 mL of SiO₂, 0–1.5% MeOH in CH₂Cl₂ gradient elution) afforded 119 (18.7 mg, 0.0433 mmol, 83%, >19:1 dr) as a colorless oil: [α]²⁵ₑ D +2.1 (c 0.43, CH₂Cl₂); ¹H NMR (CDCl₃, 500 MHz) δ 7.40−7.30 (m, 5H), 7.14 (br s, 1H), 5.40 (br s, 1H), 5.15 (s, 2H), 4.44–4.37 (m, 1H), 4.11–4.02 (m, 1H), 4.00–3.91 (m, 2H), 3.46–3.39 (m, 1H), 2.22–2.12 (m, 5H), 1.86–1.76 (m, 1H), 1.07–0.91 (m, 9H); ¹³C NMR (CDCl₃, 125 MHz) δ 168.4, 164.0, 156.7, 153.5, 136.0, 128.6 (2C), 128.3, 128.2 (2C), 119.4, 67.4, 67.1, 66.1, 44.8, 29.9, 28.8, 19.4, 18.7, 18.2, 11.4; IR (film) νmax 3313, 2967, 2100, 1721, 1509, 1212 cm⁻¹; HRMS (ESI) m/z 432.2118 (MH⁺, C₂₁H₂₉N₅O₅H⁺ requires 432.2247).

Benzyl (R,Z)-(2-((1-(1-hydroxy-3-methylbutan-2-yl)amino)-3-methyl-1-oxopent-2-en-2-yl)amino)-2-oxoethyl)carbamate (120). A solution of azide 119 (8.7 mg, 0.020 mmol) in THF (550 μL) and H₂O (90 μL) at 0 °C under Ar was treated dropwise with PMe₃ (1 M in THF, 61 μL, 0.061 mmol, 3.0 equiv). The resulting mixture was stirred at 0 °C to rt for 22 h, at which time the starting material had disappeared as evidenced by MS. The mixture was treated dropwise with piperidine (92 μL, 79 mg, 0.93 mmol), and stirred at rt for 24 h followed by concentration in vacuo. The residue was dissolved in EtOAc (10 mL), washed with H₂O (2 × 2 mL) and brine (2 mL), dried (Na₂SO₄), and concentrated in vacuo. Flash
chromatography (3 mL of SiO$_2$, 0–3% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 120 (5.5 mg, 0.014 mmol, 67%, 10:1 dr) as a white film: $[\alpha]_{D}^{25} +27$ (c 0.033, CHCl$_3$); $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.51 (br s, 1H), 7.41–7.31 (m, 5H), 6.23 (br s, 1H), 5.43 (br s, 1H), 5.18–5.09 (m, 2H), 3.95–3.84 (m, 2H), 3.77 (br s, 2H), 3.63–3.56 (m, 1H), 3.15 (br s, 1H), 2.06 (q, $J = 7.2$ Hz, 2H), 1.97 (s, 3H), 1.92–1.83 (m, 1H), 1.03–0.91 (m, 9H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 169.2, 167.3, 157.1, 139.8, 135.9, 128.6 (2C), 128.4, 128.1 (2C), 124.3, 67.5, 63.2, 57.8, 44.8, 29.0, 26.5, 19.7, 19.1, 17.8, 11.6; IR (film) $\nu_{\text{max}}$ 3289, 2924, 2360, 1654, 1522, 1255, 1147 cm$^{-1}$; HRMS (ESI) $m/z$ 406.2322 (MH$^+$, C$_{21}$H$_{31}$N$_3$O$_5$H$^+$ requires 406.2342).

2-Azidoethyl (2S*,3S*)-2-(2-((benzyloxy)carbonyl)amino)-3-hydroxy-3-methylpentanoate (121a). A solution of ester 80 (163.4 mg, 0.446 mmol) in $t$-BuOH–H$_2$O (3:1, 1.6 mL) at 0 °C was treated with LiOH•H$_2$O (93.1 mg, 2.22 mmol, 5.0 equiv), then stirred at 0 °C for 2 h. The resulting mixture was acidified to pH 1~2 by the addition of 2 N HCl (2 mL) and extracted with EtOAc (6 × 3 mL). The combined organic layers were dried (Na$_2$SO$_4$) and concentrated in vacuo. The crude carboxylic acid (152.2 mg, 150.9 mg theoretical yield, quant.) was used directly without further purification.

A solution of the crude carboxylic acid (38.3 mg, 0.113 mmol) and iodide 78a (47.7 mg, 0.242 mmol, 2.1 equiv) in anhydrous DMF (1.1 mL) at rt under Ar was treated with Et$_3$N (49 µL, 36 mg, 0.35 mmol, 3.1 equiv). The resulting mixture was stirred at 80 °C under Ar for 48 h, then concentrated in vacuo. The residue was dissolved in EtOAc (20 mL) and washed with brine (3 × 3 mL). The organic layer was dried (Na$_2$SO$_4$) and concentrated in vacuo. Flash chromatography (3 mL of SiO$_2$, 0–2% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 121a (43.6 mg, 0.107 mmol,
95% from 80) as a colorless oil: \(^1\)H NMR (CDCl\(_3\), 500 MHz) \(\delta\) 7.38–7.30 (m, 5H), 6.92 (d, \(J = 8.2\) Hz, 1H), 5.51 (s, 1H), 5.13 (s, 2H), 4.60 (d, \(J = 8.9\) Hz, 1H), 4.34–4.24 (m, 2H), 3.97–3.89 (m, 2H), 3.56–3.46 (m, 2H), 2.50 (br s, 1H), 1.55 (q, \(J = 7.2\) Hz, 2H), 1.16 (s, 3H), 0.99 (t, \(J = 7.4\) Hz, 3H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \(\delta\) 170.9, 169.2, 156.6, 136.1, 128.6 (2C), 128.3 (2C), 128.1, 74.3, 67.3, 63.7, 58.7, 49.5, 44.5, 32.4, 22.7, 8.0; IR (film) \(\nu_{\text{max}}\) 3353, 2925, 2106, 1728, 1522, 1259, 1050 cm\(^{-1}\); HRMS (ESI) \(m/z\) 408.1836 (MH\(^+\), \(\text{C}_{18}\text{H}_{25}\text{N}_{5}\text{O}_{6}\)H\(^+\) requires 408.1883).

(R)-2-Azido-3-methylbutyl \((2S^*,3S^*)\)-2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-hydroxy-3-methylpentanoate (121b). A solution of the acid derived from hydrolysis of ester 80 (prepared as described for azidoethyl ester 121a, 30.7 mg, 0.0907 mmol) and iodide 78b (44 mg, 0.184 mmol, 2.0 equiv) in anhydrous DMF (870 \(\mu\)L) at rt under Ar was treated with Et\(_3\)N (38 \(\mu\)L, 28 mg, 0.27 mmol, 3.0 equiv). The resulting mixture was stirred at 80 °C under Ar for 24 h, then the reaction mixture was dissolved in EtOAc (20 mL) and washed with brine (3 \(\times\) 3 mL). The organic layer was dried (Na\(_2\)SO\(_4\)) and concentrated in vacuo. Flash chromatography (5 mL of SiO\(_2\), 0–1.5% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 121b (34.8 mg, 0.0774 mmol, 85% from 80) as a yellow oil that was a 1:1 mixture of diastereomers: \(^1\)H NMR (CDCl\(_3\), 500 MHz) \(\delta\) 7.40–7.29 (m, 5H), 6.85 and 6.83 (2d, \(J = 10.6\) Hz, 1H), 5.44 (s, 1H), 5.13 (s, 2H), 4.62 and 4.61 (2d, \(J = 9.6\) Hz, 1H), 4.39–4.27 (m, 1H), 4.19–4.08 (m, 1H), 3.98–3.88 (m, 2H), 3.48–3.43 and 3.43–3.37 (2m, 1H), 2.44 and 2.40 (2s, 1H), 1.92–1.79 (m, 1H), 1.62–1.50 (m, 2H), 1.17 (s, 3H), 1.03–0.95 (m, 9H); \(^{13}\)C NMR (CDCl\(_3\), 125 MHz) \(\delta\) 171.0 and 170.7, 169.1 and 169.0, 156.6, 136.1, 128.6 (2C), 128.3 (2C), 128.2, 74.3, 67.3, 66.7 and 66.6, 66.4 and 66.1, 58.7, 44.5, 32.4, 30.2 and 30.0, 22.7 and 22.6, 19.4, 18.1, 8.0; IR (film)
ν_{max} 3344, 2968, 2101, 1735, 1522, 1262 cm^{-1}; HRMS (ESI) m/z 450.2345 (MH^{+}, C_{21}H_{31}N_{5}O_{6}H^{+}
requires 450.2353).

2-Azidoethyl (E)-2-2-(((benzyl)carbonyl)amino)acetamido)-3-methylpent-2-enoate (122a). A solution of alcohol 121a (19.3 mg, 0.0474 mmol) in anhydrous CHCl_{3} (210 \mu L) was treated with Martin sulfurane (0.24 M in anhydrous CHCl_{3}, 400 \mu L, 0.096 mmol, 2.0 equiv) dropwise at 0 °C under Ar. The resulting mixture was stirred at 0 °C for 1 h and concentrated in vacuo. Flash chromatography (3 mL of SiO_{2}, 0–2% MeOH in CH_{2}Cl_{2} gradient elution) afforded 122a (13.8 mg, 0.0354 mmol, 75%, >19:1 dr) as a light yellow oil: \textsuperscript{1}H NMR (CDCl_{3}, 500 MHz) δ 7.39–7.30 (m, 5H), 7.16 (s, 1H), 5.38 (s, 1H), 5.15 (s, 2H), 4.30 (t, J = 5.0 Hz, 2H), 3.95 (d, J = 5.9 Hz, 2H), 3.47 (t, J = 4.6 Hz, 2H), 2.55 (q, J = 7.5 Hz, 2H), 1.81 (s, 3H), 1.12 (t, J = 7.5 Hz, 3H); \textsuperscript{13}C NMR (CDCl_{3}, 125 MHz) δ 167.7, 163.5, 156.7, 152.6, 136.0, 128.6 (2C), 128.4, 128.2 (2C), 119.7, 67.4, 63.4, 49.8, 44.8, 27.7, 19.8, 12.6; IR (film) ν_{max} 3313, 2936, 2107, 1722, 1515, 1264 cm^{-1}; HRMS (ESI) m/z 390.1776 (MH^{+}, C_{18}H_{23}N_{5}O_{5}H^{+}
requires 390.1777).

(R)-2-azido-3-methylbutyl (E)-2-2-(((benzyl)carbonyl)amino)acetamido)-3-methylpent-2-enoate (122b). A solution of alcohol 121b (24.5 mg, 0.0545 mmol) in anhydrous CHCl_{3} (240 \mu L) was treated with Martin sulfurane (0.24 M in anhydrous CHCl_{3}, 460 \mu L, 0.11 mmol, 2.0 equiv) dropwise at –20 °C. The resulting mixture was stirred at –20 °C under Ar for 1 h, warmed to rt, and concentrated in vacuo. Flash chromatography (10 mL of SiO_{2}, 0–1.5% MeOH in CH_{2}Cl_{2} gradient elution) afforded 122b (19.7 mg, 0.0457 mmol,
84%, >19:1 dr) as a colorless oil: $[\alpha]^{25}_D +3.7$ (c 0.30, CH$_2$Cl$_2$); $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.40–7.30 (m, 5H), 7.17 (br s, 1H), 5.40 (br s, 1H), 5.15 (s, 2H), 4.44–4.37 (m, 1H), 4.12–4.01 (m, 1H), 4.00–3.89 (m, 2H), 3.47–3.40 (m, 1H), 2.57 (qd, $J = 7.5$, 1.9 Hz, 2H), 1.88–1.72 (m, 4H), 1.12 (t, $J = 7.4$ Hz, 3H), 0.98 (d, $J = 6.9$ Hz, 3H), 0.96 (d, $J = 6.8$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 167.7, 163.6, 156.7, 153.1, 136.0, 128.6 (2C), 128.3, 128.2 (2C), 119.6, 67.4, 67.0, 66.1, 44.8, 29.9, 27.7, 19.9, 19.4, 18.2, 12.6; IR (film) $\nu$$_{max}$ 3321, 2966, 2101, 1725, 1514, 1264 cm$^{-1}$; HRMS (ESI) $m/z$ 432.2261 (MH$^+$, C$_{21}$H$_{29}$N$_5$O$_5$H$^+$ requires 432.2247).

Benzyl (R,E)-(2-((1-(1-hydroxy-3-methylbutan-2-yl)amino)-3-methyl-1-oxopent-2-en-2-yl)amino)-2-oxoethyl)carbamate (123). A solution of azide 122b (6.1 mg, 0.014 mmol) in DMF (400 μL) and H$_2$O (31 μL) at 0 °C under Ar was treated dropwise with PMe$_3$ (1 M in THF, 43 μL, 0.043 mmol, 3.0 equiv). The resulting mixture was stirred at 0 °C to rt for 24 h, at which time the starting material had disappeared as evidenced by MS. The mixture was then treated dropwise with morpholine (173 μL, 174 mg, 2.0 mmol) and stirred at rt for 80 h followed by concentration in vacuo. The residue was dissolved in EtOAc (10 mL), washed with H$_2$O (2 × 2 mL) and brine (2 mL), dried (Na$_2$SO$_4$), and concentrated in vacuo. Flash chromatography (3 mL of SiO$_2$, 0–3% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 123 (5.4 mg, 0.013 mmol, 94%, 13:1 dr) as a white film: $[\alpha]^{25}_D +38$ (c 0.12, CHCl$_3$); $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.44–7.31 (m, 6H), 6.15 (d, $J = 5.0$ Hz, 1H), 5.39 (br s, 1H), 5.19–5.09 (m, 2H), 3.97–3.85 (m, 2H), 3.84–3.76 (m, 2H), 3.65–3.57 (m, 1H), 3.16 (br s, 1H), 2.36 (q, $J = 7.5$ Hz, 2H), 1.94–1.84 (m, 1H), 1.69 (s, 3H), 1.09 (t, $J = 7.5$ Hz, 3H), 0.98 (d, $J = 6.7$ Hz, 3H), 0.97 (d, $J = 6.8$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 168.7, 166.8, 157.1, 139.0, 135.8, 128.6 (2C), 128.4, 128.2 (2C), 124.8, 67.6, 63.2, 57.8, 44.9, 29.0, 27.0, 19.7, 19.0, 16.8, 12.9; IR (film) $\nu$$_{max}$ 3284,
2923, 2360, 1653, 1522, 1232, 1048 cm⁻¹; HRMS (ESI) m/z 406.2338 (MH⁺, C₂₁H₃₁N₃O₂H⁺ requires 406.2342).

[R]-2-azido-1-iodopropane (78c). A solution of (R)-(−)-2-amino-1-propanol (222 µL, 214 mg, 2.85 mmol), K₂CO₃ (386.7 mg, 2.80 mmol), and CuSO₄•5H₂O (7.7 mg, 0.031 mmol) in H₂O (9 mL) and MeOH (18 mL) at rt under Ar was treated with a solution of TfN₃ in CH₂Cl₂ (prepared according to the procedure of Lundquist and Pelletier, ca. 0.42 M, 13.3 mL, ca. 5.6 mmol). The resulting mixture was stirred at rt for 48 h. The layers were separated, and the aqueous layer was extracted with CH₂Cl₂ (4 × 20 mL). The combined organic layers were washed with brine (20 mL), dried (Na₂SO₄), and concentrated in vacuo. The crude azido alcohol was used in the next step without further purification.

A solution of PPh₃ (931 mg, 3.55 mmol) in anhydrous CH₂Cl₂ (30 mL) at rt under Ar was treated with imidazole (520 mg, 7.64 mmol) followed by I₂ (1.44 g, 5.67 mmol), stirred for 5 min, then treated dropwise with the crude azido alcohol. The resulting mixture was refluxed for 48 h, cooled to rt, and treated with sat aq Na₂SO₃ (20 mL). It was stirred until the color changed from black to yellow, at which time the layers were separated. The aqueous layer was extracted with CH₂Cl₂ (4 × 20 mL), and the combined organic layers were washed with brine (20 mL), dried (Na₂SO₄), and concentrated in vacuo. Flash chromatography (100 mL of SiO₂, 0–0.5% EtOAc in hexanes gradient elution) afforded 78c (449 mg, 2.13 mmol, 74% over 2 steps) as a light yellow oil: [α]₂⁵_D −29 (c 0.90, CHCl₃); ¹H NMR (CDCl₃, 500 MHz) δ 3.65–3.56 (m, 1H), 3.28–3.18 (m, 2H), 1.38 (d, J = 6.5 Hz, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 57.8, 19.9, 9.7; IR (film) n_max 2922, 2851, 2105, 1261 cm⁻¹.
(R)-2-Azidopropyl (2S*,3R*)-2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-hydroxy-3-methylpentanoate (124). A solution of the acid derived from hydrolysis of ester 77 (prepared as described for azidoethyl ester 114a, 6.3 mg, 0.019 mmol) and iodide 78c (10.5 mg, 0.0498 mmol) in anhydrous DMF (0.13 mL) at rt under Ar was treated with Et₃N (10 µL, 7.3 mg, 0.072 mmol). The resulting mixture was stirred at 80 °C under Ar for 24 h, then treated with additional iodide 78c (6.0 mg, 0.028 mmol) and Et₃N (6.0 µL, 4.4 mg, 0.043 mmol) and stirred at 80 °C for 24 h. It was then concentrated in vacuo and purified by flash chromatography (1.5 mL of SiO₂, 0–2% MeOH in CH₂Cl₂ gradient elution) to afford 124 (6.2 mg, 0.015 mmol, 79%, 77% from 77) as a light yellow oil that was a 1:1 mixture of diastereomers: ¹H NMR (CDCl₃, 500 MHz) δ 7.37–7.30 (m, 5H), 6.73 (s, 1H), 5.36 (s, 1H), 5.14 (s, 2H), 4.60–4.57 (m, 1H), 4.21–4.16 (m, 1H), 4.11–4.04 (m, 1H), 3.97–3.88 (m, 2H), 3.81–3.74 (m, 1H), 2.32 (s, 1H), 1.54 (q, J = 7.3 Hz, 2H), 1.30–1.22 (m, 6H), 0.92 (t, J = 7.1 Hz, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 170.8 and 170.7, 169.1, 156.5, 136.1, 128.6 (2C), 128.3 (2C), 128.1, 73.9, 68.1 and 67.9, 67.3, 58.3 and 58.2, 55.7 and 55.6, 44.5, 31.6, 23.5, 16.0 and 15.9, 7.9; IR (film) n max 3345, 2920, 2121, 1730, 1671, 1523, 1259, 1156, 1051 cm⁻¹; HRMS (ESI) m/z 422.2104 (MH⁺, C₁₉H₂₇N₅O₆H⁺ requires 422.2040).

(R)-2-azidopropyl (Z)-2-(2-(((benzyloxy)carbonyl)amino)acetamido)-3-methylpent-2-enoate (125). A solution of alcohol 124 (6.0 mg, 0.0142 mmol) in anhydrous CHCl₃ (70 µL) was treated with Martin sulfurane (0.21 M in anhydrous CHCl₃, 140 µL, 0.029 mmol, 2.0 equiv) dropwise at rt under Ar. The resulting mixture was stirred at 50 °C for 1 h and
concentrated in vacuo. Flash chromatography (5 mL of SiO$_2$, 0–2.5% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 125 (5.1 mg, 0.013 mmol, 89%, >19:1 dr) as a light yellow oil: \([a]^{25}_D −36 (c 0.50, \text{CHCl}_3)\); $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.39–7.30 (m, 5H), 7.16 (s, 1H), 5.42 (s, 1H), 5.15 (s, 2H), 4.23 (dd, $J = 11.6$, 3.5 Hz, 1H), 4.01 (dd, $J = 11.4$, 7.6 Hz, 1H), 3.96 (d, $J = 5.9$ Hz, 2H), 3.78–3.70 (m, 1H), 2.19–2.14 (m, 5H), 1.23 (d, $J = 6.7$ Hz, 3H), 1.02 (t, $J = 7.1$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 168.4, 163.9, 156.7, 153.2, 136.0, 128.6 (2C), 128.4, 128.2 (2C), 119.5, 67.7, 67.4, 56.1, 44.8, 28.7, 18.7, 16.0, 11.5; IR (film) $\nu_{\text{max}}$ 3316, 2937, 2119, 1722, 1515, 1260 cm$^{-1}$; HRMS (ESI) $m/z$ 404.1960 (MH$^+$, C$_{19}$H$_{25}$N$_5$O$_5$H$^+$ requires 404.1934).

![Chemical Structure](image)

Benzyl (R,Z)-(2-((1-(1-hydroxypropan-2-yl)amino)-3-methyl-1-oxopent-2-en-2-yl)amino)-2-oxoethyl)carbamate (126). A solution of azide 125 (6.8 mg, 0.017 mmol) in THF (460 μL) and H$_2$O (76 μL) at 0 °C under Ar was treated dropwise with PMe$_3$ (1 M in THF, 51 μL, 0.051 mmol, 3.0 equiv). The resulting mixture was stirred at 0 °C to rt for 62 h, at which time the starting material had disappeared as evidenced by MS. The mixture was then treated dropwise with piperidine (76 μL, 66 mg, 0.77 mmol) and stirred at rt for 30 h followed by concentration in vacuo after adding 0.4 mL DMF. The residue was dissolved in EtOAc (10 mL), washed with H$_2$O (2 × 2 mL) and brine (2 mL), dried (Na$_2$SO$_4$), and concentrated in vacuo. Flash chromatography (3 mL of SiO$_2$, 0–3% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 126 (5.5 mg, 0.015 mmol, 86%, 6:1 dr) as a white film: $^1$H NMR (CDCl$_3$, 500 MHz) $\delta$ 7.51 (s, 1H), 7.40–7.30 (m, 5H), 6.27 (br s, 1H), 5.47 (br s, 1H), 5.13 (s, 2H), 4.11 (br s, 1H), 3.93–3.83 (m, 2H), 3.78–3.72 (m, 1H), 3.48–3.41 (m, 1H), 3.21 (br s, 1H), 2.05 (q, $J = 7.5$ Hz, 2H), 1.99 (s,
3H), 1.18 (d, $J = 7.0$ Hz, 3H), 0.99 (t, $J = 7.5$ Hz, 3H); HRMS (ESI) $m/z$ 378.2020 (MH$^+$, C$_{19}$H$_{27}$N$_3$O$_5$H$^+$ requires 378.2029).

(R)-2-Azidopropyl (2$S^*$,3$S^*$)-2-(2-(((benzyl)oxy)carbonyl)amino)acetamido)-3-hydroxy-3-methylpentanoate (127). A solution of the acid derived from hydrolysis of ester 80 (prepared as described for azidoethyl ester 121a, 25.0 mg, 0.0739 mmol) and iodide 78c (93.8 mg, 0.445 mmol, 6.0 equiv) in anhydrous DMF (750 mL) at rt under Ar was treated with Et$_3$N (83 µL, 60 mg, 0.60 mmol, 8.1 equiv). The resulting mixture was stirred at 75 °C under Ar for 72 h, then concentrated in vacuo. The residue was dissolved in EtOAc (10 mL) and washed with brine (2 × 2 mL). The organic layer was dried (Na$_2$SO$_4$) and concentrated in vacuo. Flash chromatography (5 mL of SiO$_2$, 0–3% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 127 (17.1 mg, 0.0406 mmol, 55% from 80) as a light yellow oil that was a 1:1 mixture of diastereomers. $^1$H NMR (CDCl$_3$, 500 MHz) δ 7.38–7.30 (m, 5H), 6.78 (s, 1H), 5.38 (s, 1H), 5.14 (s, 2H), 4.62–4.58 (m, 1H), 4.22–4.14 (m, 1H), 4.10–4.04 (m, 1H), 3.98–3.88 (m, 2H), 3.81–3.74 (m, 1H), 2.31 (br s, 1H), 1.55 (q, $J = 6.9$ Hz, 2H), 1.28 (t, $J = 7.4$ Hz, 3H), 1.16 (s, 3H), 0.99 (t, $J = 7.4$ Hz, 3H); $^{13}$C NMR (CDCl$_3$, 125 MHz) δ 170.8 and 170.7, 169.0, 156.5, 136.0, 128.6 (2C), 128.3 (2C), 128.2, 74.3, 68.0 and 67.8, 67.3, 58.7, 55.7 and 55.6, 44.5, 32.5, 22.6, 16.0 and 15.9, 8.0; IR (film) $n_{max}$ 3341, 2977, 2921, 1731, 1673, 1524, 1261, 1051 cm$^{-1}$; HRMS (ESI) $m/z$ 422.2025 (MH$^+$, C$_{19}$H$_{27}$N$_5$O$_6$H$^+$ requires 422.2040).

(R)-2-azidopropyl (E)-2-(((benzyl)oxy)carbonyl)amino)acetamido)-3-methylpent-2-enoate (128). A solution of alcohol 127 (10.1 mg, 0.0240 mmol) in anhydrous
CHCl₃ (150 μL) was treated with Martin sulfurane (0.32 M in anhydrous CHCl₃, 150 μL, 0.051 mmol, 2.1 equiv) dropwise at rt under Ar. The resulting mixture was stirred at 50 °C for 1 h and concentrated in vacuo. Flash chromatography (5 mL of SiO₂, 0–2.5% MeOH in CH₂Cl₂ gradient elution) afforded 128 (8.1 mg, 0.020 mmol, 84%, >19:1 dr) as a light yellow oil: [α]²⁵⁰ D −30 (c 0.40, CHCl₃); ¹H NMR (CDCl₃, 500 MHz) δ 7.38–7.30 (m, 5H), 7.15 (s, 1H), 5.39 (s, 1H), 5.16 (s, 2H), 4.24 (dd, J = 11.6, 3.5 Hz, 1H), 4.01 (dd, J = 11.5, 7.8 Hz, 1H), 3.95 (d, J = 5.9 Hz, 2H), 3.97–3.93 (m, 2H), 3.81–3.71 (m, 1H), 2.56 (q, J = 7.5 Hz, 2H), 1.82 (s, 3H), 1.23 (d, J = 6.7 Hz, 3H), 1.12 (t, J = 7.5 Hz, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 167.7, 163.5, 156.7, 152.9, 136.0, 128.6 (2C), 128.4, 128.2 (2C), 119.7, 67.7, 67.4, 56.1, 44.8, 27.7, 19.8, 16.0, 12.7; IR (film) νₘₐₓ 3314, 2920, 2850, 2120, 1723, 1514, 1264, 1207 cm⁻¹; HRMS (ESI) m/z 404.1949 (MH⁺, C₁₉H₂₅N₅O₅H⁺ requires 404.1934).

**Ethyl (R*)-3-hydroxy-3-methyl-2-(((R)-2,2,2-trichloro-1-phenylethoxy)carbonyl)amino)butanoate (148).** A solution of (R)-2,2,2-trichloro-1-phenylethyl ((methylsulfonyl)oxy)carbamate 147 (740 mg, 2.04 mmol, 1.2 equiv) in CH₃CN (10 mL) at rt was treated with OsO₄ (4 wt % solution in H₂O, 0.6 mL, 0.094 mmol, 0.075 equiv), stirred at rt for 20 min, then treated with the Ethyl 3-methylbut-2-enoate 54 (159 mg, 1.24 mmol, 1 equiv) and H₂O (0.68 ml). The resulting mixture was stirred at 40 °C for 48 h, then worked up following standard procedure. Flash chromatography (30 mL SiO₂, 0–1% MeOH in CH₂Cl₂ gradient elution) afforded 151(348.1 mg, 0.843 mmol, 68%) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz, consisted of 2 diastereomers) δ 7.62 (d, J = 6.5 Hz, 2H), 7.46–7.35 (m, 3H), 6.33 (s, 0.5H), 6.27 (s, 0.5H), 6.02–5.93 (m, 1H), 4.43–4.11 (m, 3H), 2.59–2.47 (m, 1H), 1.45–1.16 (m, 6H), 1.23 (s, 3H); ¹³C NMR (CDCl₃, 125 MHz) δ 171.3, 171.2, 154.3,154.2, 133.1, 133.0, 129.7
(2C), 129.6, 127.9 (2C), 99.7, 99.4, 83.7, 83.5, 72.1, 71.7, 61.8 (2C), 61.6, 61.5, 27.0, 26.3, 26.2, 14.1, 14.0; HRMS (ESI) m/z 412.0503 (MH⁺, C₁₆H₂₀Cl₃NO₅H⁺ requires 412.0485).

Ethyl (R)-3-methyl-2-(((R)-2,2,2-trichloro-1-phenylethoxy)carbonyl)amino)-3-((triethylsilyloxy)butanoate (151) and Ethyl (S)-3-((ethyl-l₂-silyloxy)-3-methyl-2-(((R)-2,2,2-trichloro-1-phenylethoxy)carbonyl)amino)butanoate (152). A solution of 1:1 diastereomeric 148 (120 mg, 0.291 mmol, 1.0 equiv) in CH₂Cl₂ (4 mL) at 0 °C was treated with 2, 6-lutidine (0.1 ml, 0.882 mmol, 3.0 equiv) followed by TES-OTf (0.13 ml, 0.577 mmol, 2.0 equiv). The resulting mixture was stirred at 0 °C for 2 h, then treated with H₂O and and extracted with CH₂Cl₂ (3 times), dried (Na₂SO₄), and concentrated in vacuo. Simple column separation (5 mL SiO₂ at 50% hexane–CH₂Cl₂) delivered the desired product (160 mg, quant.). For convenience, the crude material is normally subjected to flash chromatography (SiO₂) to separate the diastereomers. 1 g of the crude 1:1 mixture after 3 chromatographic separation (40 mL of SiO₂, 10%–50% CH₂Cl₂ in hexane gradient elution) can afford 100 mg 151 and 100 mg 152 each with dr>10:1 as colorless oils. For 151: ¹H NMR (CDCl₃, 500 MHz) δ 7.63 (d, J = 7.5 Hz, 2H), 7.44–7.36 (m, 3H), 6.32 (s, 1H), 5.85 (d, J = 9.5 Hz, 1H), 4.19–4.09 (m, 3H), 1.39 (s, 3H), 1.35 (s, 3H), 0.98 (t, J = 7.5 Hz, 9H), 0.62 (J, J = 8.0 Hz, 6H); ¹³C NMR (CDCl₃, 125 MHz) δ 169.9, 153.9, 133.2, 129.7, 129.6 (3C), 127.9, 99.8, 83.3, 75.0, 63.1, 27.7, 27.6, 14.0, 7.0, 6.9, 6.5. For 152: ¹H NMR (CDCl₃, 500 MHz, major rotamor) δ 7.62 (d, J = 6.5 Hz, 2), 7.44–7.35 (m, 3H), 6.27 (s, 1H), 5.83 (d, J = 9.0 Hz, 1H), 4.27–4.17 (m, 2H), 4.11 (d, J = 9.5Hz, 1H), 1.35 (s, 3H), 1.30 (t, J = 7.0 Hz, 3H), 1.21 (s, 3H), 0.95 (t, J = 8.0 Hz, 9H), 0.58 (q, J = 8.0 Hz, 6H); ¹³C NMR (CDCl₃, 125 MHz) δ 169.8, 154.0,
HRMS (ESI) $m/z$ 526.1386 (MH$^+$, C$_{22}$H$_{34}$Cl$_3$NO$_5$SiH$^+$ requires 526.1350).

Ethyl (2R*,3S*)-3-hydroxy-3-methyl-2-(((R)-3-methyl-2-(((R)-2,2,2-trichloro-1-phenylethoxy)carbonyl)amino)-3-((triethylsilyl)oxy)butanamido)pentanoate (164). A suspension of 151 (106.3 mg, 0.202 mmol) and Me$_3$SnOH (80.0 mg, 0.444 mmol, 2.2 equiv) in hexane (8 mL, pretreated with Na$_2$SO$_4$ for 6 h) was flushed with Ar and stirred at 60 °C for 48 h. The solvent was evaporated, and 10 mL diethyl ether was added. The mixture was filtered through Celite, and the Celite pad was washed with diethyl ether (60 mL). The filtrate was concentrated in vacuo to afford 163 as a colorless oil, which was used directly in the next step without further purification.

A solution of the acid in anhydrous CH$_2$Cl$_2$ (4 mL) at 0 °C under Ar was treated with amine 153 (49.6 mg, 0.283 mmol, 1.4 equiv), HOBt (ca. 20% H$_2$O content, 51.0 mg, 0.310 mmol, 1.54 equiv), and EDC•HCl (58.0 mg, 0.303 mmol, 1.5 equiv). The resulting mixture stirred at 0 °C under Ar for 3 h. The reaction was quenched by the addition of sat. aq, NaHCO$_3$ (5 mL) and the aqueous layer was extracted with CH$_2$Cl$_2$ ($\times$ 5 mL), dried (Na$_2$SO$_4$), and concentrated in vacuo. Flash chromatography (10 mL of SiO$_2$, 0–1.5% MeOH in CH$_2$Cl$_2$ gradient elution) afforded 77 (111.0 mg, 0.170 mmol, 84%) as a colorless oil: $^1$H NMR (CDCl$_3$, 500 MHz, consisted of two diastereomers) $\delta$ 7.61 (d, $J = 7.5$ Hz, 2H), 7.44–7.34 (m, 3H), 6.32 (s, 1H), 6.11 (d, $J = 8.0$ Hz, 0.5H), 6.05 (d, $J = 7.5$ Hz, 0.5H), 4.55 (d, $J = 8.5$ Hz, 0.5H), 4.41 (d, $J = 8.5$ Hz, 0.5H), 4.27–4.13 (m, 3H), 2.55 (s, 0.5 H), 2.50 (s, 0.5 H), 1.58–1.45 (m, 2H), 1.44–1.39 (m, 3H), 1.34–1.24 (m, 6H), 1.21–1.17 (m, 3H), 1.03–0.97 (m, 9H), 0.87 (t, $J = 7.5$ Hz, 3H), 0.76–0.66 (m, 6H); $^{13}$C NMR (CDCl$_3$, 125 MHz) $\delta$ 171.7, 171.1, 170.0, 169.6, 154.7, 154.5, 133.1, 129.7 (3C),
129.6, 127.9, 99.8, 99.7, 83.4, 76.1, 74.1, 73.4, 63.7, 62.8, 61.6, 61.5, 58.5, 58.4, 31.6, 31.3, 27.5, 27.3, 26.0, 25.0, 23.7, 23.3, 14.2, 14.1, 7.9, 7.7, 6.8, 6.5. IR (film) νmax 3358, 2957, 2877, 2359, 1736, 1665, 1508, 1372, 1203, 1057 cm⁻¹; HRMS (ESI) m/z 655.2134 (MH⁺, C₁₈H₂₆N₂O₆H⁺ requires 655.2140).

**Ethyl (2R*,3S*)-2-((R)-2-((tert-butoxycarbonyl)amino)-3-methyl-3-((triethylsilyl)oxy)butanamido)-3-hydroxy-3-methylpentanoate (166).** A suspension of 164 (53.0 mg, 0.081 mmol) in THF/Sat. aq NaHCO₃ 3:1 solution (4 mL) was treated with 10% Pd/C (9.6 mg, 0.18 wt eqiv) and Boc₂O (18.5 mg, 0.085 mmol, 1.05 equiv.) sequentially at rt under Ar, and stirred at rt under H₂ (100 psi) for 15 h. H₂O (2 mL) were added and extracted with EtOAc (5 × 3 mL). The combined organic extracts were dried (Na₂SO₄), and concentrated in vacuo. Flash chromatography (3.5 mL of SiO₂, 0–1% MeOH in CH₂Cl₂ gradient elution) afforded 166 (37.3 mg, 0.074 mmol, 92%) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz) 7.14 (s, 1H), 5.45 (br s, 1H), 4.62 (d, J = 8.5 Hz, 1H), 4.30–4.17 (m, 2H), 4.04 (br s, 1H), 2.75 (s, 1H), 2.65 (s, 0.3H), 1.57 (q, J = 7.5 Hz, 2H), 1.45 (s, 9H), 1.39–1.25 (m, 9H), 1.18 (s, 3H), 1.03–0.89 (m, 12H), 0.73–0.64 (m, 6H); ¹³C NMR (CDCl₃, 125 MHz) δ 171.3, 170.9, 170.5, 156.1, 80.0, 75.5, 74.2, 73.7, 63.7, 61.5, 58.5, 58.2, 31.5, 31.4, 28.3, 27.6, 27.2, 26.7, 23.6, 14.2, 14.1, 7.9, 7.0, 6.5. IR (film) νmax 3406, 2977, 2360, 1734, 1507, 1162, 1028 cm⁻¹; HRMS (ESI) m/z 505.3335 (MH⁺, C₁₈H₂₆N₂O₆H⁺ requires 505.3309).
**Tert-butyl ((6R,12R,Z)-9-(butan-2-ylidene)-3,3-diethyl-13-hydroxy-5,5,12-trimethyl-7,10-dioxo-4-oxa-8,11-diaza-3-silatridecan-6-yl)carbamate (169).** 6182  A solution of alcohol 168 (10.6 mg, 0.0189 mmol) in anhydrous CHCl₃ (100 µL) was treated with...
Martin sulfurane (25.4 mg in anhydrous 400 μL CHCl₃, 0.0378 mmol, 2.0 equiv) dropwise at 0 °C. The resulting mixture was stirred at 0 °C under Ar for 1 h, warmed to rt, and concentrated *in vacuo*. This oil mixture was treated with DMF (550 μL), H₂O (90 μL) and lindlar catalyst (100 mg) sequentially at rt. Then, the resulting suspension was stirred at rt under H₂ (1 atm) for 15 h (azide reduction can be evidenced by MS), then peperidine (90 μL) was added under Ar, and stirred at rt for 24 h. The mixture was treated with sat. aq NaHCO₃ 2 mL, and extracted with EtOAc (5 × 2 mL). The combined organic extracts were dried (Na₂SO₄), and concentrated *in vacuo*. Flash chromatography (5 mL of SiO₂, 0–4% MeOH in CH₂Cl₂ gradient elution) afforded **169** (8.3 mg, 0.0161 mmol, 75%, 12:1 dr) as a white film: [α]²⁵⁺D +7.2 (c 0.21, CHCl₃); ¹H NMR (CDCl₃, 500 MHz) δ 7.49 (s, 1H), 6.48 (br s, 1H), 5.49 (br s, 1H), 4.13 (d, J = 4.0 Hz, 1H), 3.93 (d, J = 5.0 Hz, 1H), 3.87–3.74 (m, 1H), 3.55–3.41 (m, 1H), 3.26 (br s, 1H), 2.18–2.08 (m, 2H), 2.04 (s, 3H), 1.46 (s, 9H), 1.36 (d, J = 9.0 Hz, 6H), 1.18 (d, J = 7.0 Hz, 3H), 1.04 (t, J = 8.0 Hz, 3H), 0.98 (t, J = 9.0 Hz, 9H), 0.66 (q, J = 8.0 Hz, 6H); ¹³C NMR (CDCl₃, 125 MHz) δ 169.9, 165.6, 156.8, 141.6, 123.7, 80.8, 74.8, 66.2, 64.9, 48.0, 28.3, 27.4, 27.1, 17.8, 16.6, 11.7, 7.0, 6.3. IR (film) νmax 3348, 2924, 2283, 1665, 1461, 1367, 1169, 1051 cm⁻¹; HRMS (ESI) m/z 516.3424 (MH⁺, C₂₅H₄₉N₃O₆SiH⁺ requires 516.3469).

![Benzyl ((5-oxo-4-(propan-2-ylidene)oxazolidin-2-yl)methyl)carbamate](image)

**Benzyl ((5-oxo-4-(propan-2-ylidene)oxazolidin-2-yl)methyl)carbamate (170)** or azlactone characterization: ¹H NMR (CDCl₃, 500 MHz) δ 7.42–7.29 (m, 5H), 5.35 (br s, 1H), 5.14 (s, 2H), 4.30 (d, J = 5.3 Hz, 2H), 2.34 (s, 3H), 2.23 (s, 3H); HRMS (ESI) m/z 289.0204 (MH⁺, C₁₅H₁₈N₂O₄H⁺ requires 289.1188).
Ethyl (6S,9R)-12-(2-hydroxypropan-2-yl)-6,9-diisopropyl-2,2-
dimethyl-4,7,10-trioxo-3-oxa-5,8,11-triazatridecan-13-oate (142a). A solution of the acid Boc-Val-Val-OH (27 mg, 0.085 mmol) in anhydrous DMF–CH₂Cl₂ (3 mL 5:1) at 0 °C under Ar was treated with amine 57a (20 mg, 0.124 mmol), HOBt (ca. 20% H₂O content, 23 mg, 0.136 mmol), and EDC•HCl (21 mg, 0.110 mmol). The resulting mixture stirred at 0 °C under Ar for 3 h. The reaction was quenched by the addition of sat aq NaHCO₃ (3 mL) and H₂O (2 mL), and CH₂Cl₂ was vacuumed off. It was extracted with EtOAc (10 × 5 mL), dried (Na₂SO₄), and concentrated in vacuo. Flash chromatography (5 mL of SiO₂, 0–50% EtOAc in CH₂Cl₂ gradient elution) afforded 142a (30.4 mg, 0.066 mmol, 77%) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz, 2 diastereomers) δ 7.12–6.95 (m, 1H), 6.77 (d, J = 7.0 Hz, 0.5H), 6.68 (d, J = 8.0 Hz, 0.5H), 5.20 (t, J = 7.0 Hz, 1H), 4.51 (q, J = 5.5 Hz, 1H), 4.45–4.40 (m, 1H), 4.30–4.17 (m, 2H), 4.01 (br s, 1H), 3.10 (s, 1H), 2.33–2.08 (m, 2H), 1.87 (s, 0.5H), 1.44 (s, 9H), 1.35–1.21 (m, 9H), 1.03–0.87 (m, 12H); ¹³C NMR (CDCl₃, 125 MHz) δ 171.9, 171.8, 171.2, 171.0, 155.8, 155.7, 80.0, 71.8, 61.6, 60.0, 58.4, 58.3, 30.8, 28.3, 26.9, 26.8, 26.7, 19.4 (2C), 19.1, 18.2, 17.7, 17.5, 14.1.; IR (film) νmax 3361, 3275, 2973, 2361, 1733, 1641, 1532, 1368, 1168 cm⁻¹; HRMS (ESI) m/z 460.3038 (MH⁺, C₁₈H₂₆N₂O₆H⁺ requires 460.3023).

Tert-butyl (4S,10R,13S)-4,10-diisopropyl-2,14-dimethyl-6,9,12-
trioxo-7-(propan-2-ylidene)-2,5,8,11-tetraazapentadecan-143-yl)carbamate (140). A solution of ester 142a (40 mg, 0.087 mmol) in t-BuOH–H₂O (2:1, 3.0 mL) at 0 °C was treated with LiOH•H₂O (20 mg, 0.476 mmol, 5.5 equiv), then stirred at 0 °C for 3 h. The resulting mixture
was acidified to pH 4~5 by the addition of 2 N HCl and diluted with H₂O 3 mL, extracted with EtOAc (8 × 5 mL). The combined organic layers were dried (Na₂SO₄) and concentrated in vacuo. The crude carboxylic acid was used directly without further purification.

A solution of the acid 142 in anhydrous DMF (0.6 mL) at rt under Ar was treated EDCI·HCl (34 mg, 0.177 mmol, 2 equiv) and stirred for 15 h. The disappearance of start material was evidenced by MS. Amine 143 HCl salt (24 mg, 0.144 mmol, 1.7 equiv) was added followed by DMF (1.4 mL, to rinse the amine·HCl off vial) and NEt₃ (50 µL), and the resulting mixture was stirred at rt for 3 h. The reaction was quenched by the addition of sat. aq NaHCO₃ (2 mL) and diluted with H₂O (20 mL). The aqueous layer was extracted with EtOAc (8 × 4 mL), dried (Na₂SO₄), and concentrated in vacuo. Flash chromatography (8 mL of SiO₂, 0–3% MeOH in EtOAc with 1% NEt₃ gradient elution) afforded peptide 140 (45.1 mg, 0.0859 mmol, 97%) as a white film: [α]²⁵°D +8.8 (c 0.69, CHCl₃); ¹H NMR (CDCl₃, 500 MHz) δ 7.65 (br s, 1H), 6.96 (br s, 1H), 6.61 (br s, 1H), 6.05 (br s, 1H), 4.31–4.21 (m, 1H), 4.09–3.98 (m, 1H), 3.75–3.63 (m, 1H), 2.66 (br s, 2H), 2.46–2.32 (m, 1H), 2.13 (s, 6H), 2.17 (s, 3H), 2.15–2.02 (m, 1H), 1.96–1.85 (m, 1H), 1.75 (s, 3H), 1.43 (s, 9H), 1.10–0.82 (m, 18H); ¹³C NMR (CDCl₃, 125 MHz) δ 173.2, 170.3, 165.4, 156.8, 141.7, 123.7, 80.3, 61.5, 59.8, 59.5, 51.2, 45.3, 30.9, 29.7, 28.1, 21.7, 20.8, 19.8, 19.5, 19.1, 18.5, 17.9; IR (film) νmax 3266, 2964, 2764, 2360, 1717, 1642, 1541, 1390, 1174 cm⁻¹; HRMS (ESI) m/z 526.3980 (MH⁺, C₂₇H₅₂N₅O₅H⁺ requires 526. 3968).

Ethyl (2R*,3S*)-3-hydroxy-3-methyl-2-((2S,3R)-3-methyl-2-(((R)-2,2,2-trichloro-1-phenylethoxy)carbonyl)amino)-3-((triethylsilyl)oxy)pentanamido)pentanoate (178). A suspension of 177 (109 mg, 0.2022 mmol)
and Me₃SnOH (100.0 mg, 0.553 mmol, 2.7 equiv) in hexane (10 mL, pretreated with Na₂SO₄ for 6 h) was flushed with Ar and stirred at 60 °C for 72 h. The solvent was evaporated, and 10 mL diethyl ether was added. The mixture was filtered through Celite, and the Celite pad was washed with diethyl ether (60 mL). The filtrate was concentrated in vacuo to afford the respective carboxylic acid (125.8 mg crude weight) as a colorless oil, which was used directly in the next step without further purification.

A solution of the acid (97 mg was used out of 125.8 mg, assuming 0.155 mmol) in anhydrous CH₂Cl₂ (2 mL) at 0 °C under Ar was treated with amine \textbf{153} (30 mg, 0.171 mmol, 1.1 equiv), HOBt (ca. 20% H₂O content, 39.2 mg, 0.232 mmol, 1.5 equiv), and EDC•HCl (44.6 mg, 0.233 mmol, 1.5 equiv). The resulting mixture stirred at 0 °C under Ar for 3 h. The reaction was quenched by the addition of sat. aq NaHCO₃ (4 mL) and the aqueous layer was extracted with CH₂Cl₂ (6 × 4 mL), dried (Na₂SO₄), and concentrated in vacuo. Flash chromatography (10 mL of SiO₂, 0–2% MeOH in CH₂Cl₂ gradient elution) afforded \textbf{178} (77.3 mg, 0.115 mmol, 74% from ester \textbf{177}, 2 step) as a colorless oil: ¹H NMR (CDCl₃, 500 MHz, consisted of two diastereomers) δ 7.62 (d, J = 7.6 Hz, 2H), 7.46–7.36 (m, 3H), 6.28 (d, J = 4.6 Hz, 1H), 6.08–5.96 (m, 1H), 4.32 (d, J = 2.2 Hz, 0.5H), 4.30 (d, J = 2.2 Hz, 0.5H), 4.28–4.16 (m, 3H), 2.80–2.42 (m, 1H), 1.64–1.47 (m, 4H), 1.40–1.19 (m, 9H), 1.18 (s, 3H), 1.06–0.90 (m, 12H), 0.78–0.61 (m, 6H); HRMS (ESI) HRMS (ESI) m/z 669.2293 (MH⁺, C₃₀H₄₈N₂O₇SiH⁺ requires 669.2296).

Ethyl (2R*,3S*)-2-((2S,3R)-2-((tert-butoxycarbonyl)amino)-3-methyl-3-((triethylsilyl)oxy)pentanamido)-3-hydroxy-3-methylpentanoate (179). A suspension of carbamate \textbf{178} (84.9 mg, 0.127 mmol) in THF/Sat. aq NaHCO₃ 2:1 solution (2.3 mL) was treated with 10% Pd/C (10 mg, 0.12 wt equiv) and Boc₂O (29 mg, 0.133 mmol, 1.05 equiv) sequentially
at rt under Ar, and stirred at rt under H\(_2\) (100 psi) for 15 h. H\(_2\)O (1 mL) and Sat. aq NaHCO\(_3\) solution (1 ml) were added and extracted with EtOAc (5 \times 3 mL). The combined organic extracts were dried (Na\(_2\)SO\(_4\)), and concentrated \textit{in vacuo}. Flash chromatography (8 mL of SiO\(_2\), 0–2% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 178 (71 mg, 0.127 mmol, quant.) as a colorless oil:

\[
\begin{align*}
\text{1H NMR (CDCl}_3, 500 MHz) \delta & \text{7.41 (br s, 1H), 6.15 (br s, 1H), 4.62, 4.55 (d, } J = 8.6 \text{ Hz, 1H),} \\
& \text{4.38–4.15 (m, 3H), 4.05 (d, } J = 6.9 \text{ Hz, 0.5 H), 3.19 (d, } J = 7.0 \text{ Hz, 0.5H), 1.65–1.50 (m, 4H),} \\
& \text{1.47 (s, 9H), 1.40–1.15 (m, 9H), 1.07–0.83 (m, 15H), 0.79–0.61 (m, 6H); HRMS (ESI) } m/z 519.3458 (MH}^+\text{, } C_{18}H_{26}N_2O_6H}^+\text{ requires 519.3466).}
\end{align*}
\]

2-azidoethyl (2R\(^*\),3S\(^*\))-2-((2S,3R)-2-((tert-butoxycarbonyl)amino)-3-methyl-3-((triethylsilyl)oxy)pentanamido)-3-hydroxy-3-methylpentanoate (180). A solution of ester 179 (21.2 mg, 0.041 mmol) in \(t\)-BuOH–H\(_2\)O (2:1, 0.9 mL) at 0 °C was treated with LiOH•H\(_2\)O (10 mg, 0.238 mmol, 5.8 equiv), then stirred at rt for 3 h. The resulting mixture was added water (1 mL) and extracted with EtOAc (1.5 mL \times 5). The combined organic layers were dried (Na\(_2\)SO\(_4\)) and concentrated \textit{in vacuo}. The crude carboxylic acid was used directly without further purification.

A solution of the crude carboxylic acid and iodide 78b (14.4 mg, 0.0731 mmol, 1.8 equiv) in anhydrous DMF (0.51 mL) at rt under Ar was treated with triethylamine (15.2 \(\mu\)L, 0.109 mmol, 3.23 equiv). The resulting mixture was stirred at 80 °C under Ar for 15 h, and was added H\(_2\)O (2 mL) and extracted with EtOAc (5 \times 2 mL). The organic layer was dried (Na\(_2\)SO\(_4\)) and concentrated \textit{in vacuo}. Flash chromatography (2 mL of SiO\(_2\), 0–2% MeOH in CH\(_2\)Cl\(_2\) gradient elution) afforded 180 (17.8 mg, 0.0318 mmol, 78% from ester 179 for 2 steps) as a colorless oil:

\[
\begin{align*}
\text{1H NMR (CDCl}_3, 500 MHz, consisted of two diastereomers) } \delta & \text{7.09 (br s, 1H), 5.46–5.28 (m,} \\
& \text{4.38–4.15 (m, 3H), 4.05 (d, } J = 6.9 \text{ Hz, 0.5 H), 3.19 (d, } J = 7.0 \text{ Hz, 0.5H), 1.65–1.50 (m, 4H),} \\
& \text{1.47 (s, 9H), 1.40–1.15 (m, 9H), 1.07–0.83 (m, 15H), 0.79–0.61 (m, 6H); HRMS (ESI) } m/z 519.3458 (MH}^+\text{, } C_{18}H_{26}N_2O_6H}^+\text{ requires 519.3466).}
\end{align*}
\]
1H), 4.53 (d, J = 8.3, 0.5H), 4.39–4.35 (m, 1H), 4.35–4.25 (m, 2H), 4.16 (d, J = 1.4 Hz, 0.5H), 3.61–3.48 (m, 2H), 1.67–1.49 (m, 4H), 1.45 (s, 9H), 3.8–3.6 (m, 6H), 1.12–0.85 (m, 15H), 0.74–0.61 (m, 6H). 560.3494 (MH⁺, C₂₅H₄₉N₅O₇SiH⁺ requires 560.3480).

2tert-butyl (5R,6S,Z)-9-(butan-2-ylidene)-3,3,5-triethyl-13-hydroxy-5-methyl-7,10-dioxo-4-oxa-8,11-diaza-3-silatridecan-6-yl)carbamate (181). A solution of alcohol 180 (15.4 mg, 0.0275 mmol) in anhydrous CHCl₃ (100 µL) was treated with Martin sulfurane (37.0 mg in anhydrous 400 µL CHCl₃, 0.055 mmol, 2.0 equiv) dropwise at 0 °C. The resulting mixture was stirred at 0 °C under Ar for 1 h, warmed to rt, and concentrated in vacuo. This oil mixture was treated with THF (550 µL), H₂O (50 µL) and lindlar catalyst (130 mg) sequentially at rt. Then, the resulting suspension was stirred at rt under H₂ (1 atm) for 15 h (azide reduction can be evidenced by MS), then peperidine (50 µL) was added under Ar, and stirred at rt for 24 h. The mixture was treated with sat. NH₄Cl 0.5 mL and H₂O 1.5 mL, and the mixture was extracted with EtOAc (7 × 2 mL). The combined organic extracts were dried (Na₂SO₄), and concentrated in vacuo. Flash chromatography (5 mL of SiO₂, 0–4% MeOH in CH₂Cl₂ gradient elution) afforded 169 (10.6 mg, 0.0205 mmol, 75%, 12:1 dr) as a white film: [α]²⁵° _D_ -4.1 (c 0.35, CHCl₃); δ ¹H NMR (CDCl₃, 500 MHz) δ 7.40 (s, 1H), 6.85 (s, 1H), 5.39 (s, 1H), 4.05 (d, J = 6.0 Hz, 1H), 3.73 (s, 2H), 3.53–3.26 (m, 3H), 2.13 (q, J = 7.5 Hz, 2H), 2.04 (s, 3H), 1.75–1.60 (m, 2H), 1.46 (s, 9H), 1.35 (s, 3H), 1.04 (t, J = 8.0 Hz, 3H), 0.99 (t, J = 8.0 Hz, 9H), 0.92 (t, J = 7.5 Hz, 4H), 0.68 (q, J = 8.0 Hz, 6H); ¹³C NMR (CDCl₃, 125 MHz) δ 170.5, 167.0, 156.6, 143.3, 123.4, 80.8, 75.7, 77.7, 61.8, 42.8, 33.2, 28.3, 27.2, 24.4, 17.8, 11.7, 8.9, 7.1, 7.0, 6.7, 6.5; IR (film) νmax 3317, 2919, 2850, 1686, 1522, 1248 cm⁻¹; HRMS (ESI) m/z 516.3480 (MH⁺, C₂₅H₄₉N₅O₇SiH⁺ requires 516.3469).
5.3 References


(12) 75 could be prepared via the procedure employed by Inoue and co-workers (*J. Am. Chem. Soc.* 2013, 135, 5467-5474.) for synthesis of the analogous vinyl iodide (listed as compound 33 in their paper), using MeOH or H2O trapping instead of I2 trapping.

(13) A hydrogenation conducted on smaller scale (141 mg of 76) at 750 psi H2 was complete within 24 h.


(15) Macleod, F.; Lang, S.; Murphy, J. A. *Synlett* 2010, 529-534.

5.4 Spectra
122a

(500 MHz, CDCl₃)