Synthesis of enantiomerically enriched \(\beta,\gamma\)-unsaturated-\(\alpha\)-amino acids

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Abstract —A variety of enantiomerically enriched \(\beta,\gamma\)-unsaturated-\(\alpha\)-amino acids are synthesized by olefination of a Cbz-protected serine aldehyde equivalent, readily prepared from serine. A cyclic ortho ester protecting group is employed to minimize racemization. The deprotected amino acids are obtained in good yield, ranging from 70–95% ee, with double-bond geometry determined by the type of Wittig reagent used. Isotopically labeled side chains are readily introduced by this procedure, and free \(\gamma\)-\(^{13}\)C-vinylglycine was prepared in 44% yield from the protected serine aldehyde synthon. © 2001 Elsevier Science Ltd. All rights reserved.

1. Introduction

\(\beta,\gamma\)-Unsaturated-\(\alpha\)-amino acids 1, otherwise known as the vinylglycines, have been isolated from a variety of natural sources.\(^1\) There is considerable interest in their biological activity, in particular their ability to act as suicide substrates or mechanistic probes of pyridoxal phosphate (PLP) dependent enzymes.\(^2\) These enzymes are a vital link in many biosynthetic pathways as they are involved in catalyzing chemical changes at the \(\alpha\)-, \(\beta\)-, or \(\gamma\)-carbons of amino acids. In addition, \(\beta,\gamma\)-unsaturated-\(\alpha\)-amino acids sometimes possess antimicrobial activity,\(^3\) can be useful synthetic intermediates,\(^4\) and can serve as conformationally restricted analogs of common amino acids for structure-activity relationship studies.

The \(\beta,\gamma\)-unsaturated-\(\alpha\)-amino acids pose a synthetic challenge, primarily due to the tendency of these compounds to isomerize to the conjugated \(\alpha,\beta\)-unsaturated derivatives. A number of routes to racemic \(\beta,\gamma\)-unsaturated-\(\alpha\)-amino acids have been reported.\(^2,5\) Racemic vinylglycine itself was first synthesized in 1974 by a Strecker synthesis,\(^6\) while a number of efficient syntheses of optically active vinylglycine have been published since 1980.\(^7\) However, most procedures rely upon the degradation of an amino acid, so isotopic labeling is difficult, and no \(\gamma\)-\(^{13}\)C-labeled compounds have been reported. More versatile procedures for preparing a number of \(\beta,\gamma\)-unsaturated-\(\alpha\)-amino acids have also appeared.\(^8\) Probably the most effective synthetic route makes use of a Wittig olefination of the serine derived Garner aldehyde 2 to generate a \(\beta,\gamma\)-unsaturated amino alcohol.\(^9a\) An oxidation step is then required to generate the amino acid, limiting the type of side chain functional groups which can be present.\(^9b-d\) A variation of this strategy employs a cysteine derived N-acylthiazolidinone as the cyclic aldehyde; much milder oxidation conditions are required to regenerate the acid moiety.\(^10\)

The \(\beta,\gamma\)-unsaturated-\(\alpha\)-amino acids have recently described the development of a new strategy for the synthesis of a variety of classes of amino acids, based upon the elaboration of an optically active serine-derived aldehyde in which the optical integrity is maintained by a cyclic ortho ester carboxyl protecting group.\(^11-14\) These syntheses have previously utilized the 9-fluorenylmethoxycarbonyl (Fmoc) group for amine protection. While this base sensitive moiety is stable to the reaction of the serine aldehyde with stabilized ylides,\(^11\) it is quickly cleaved during attempts at reaction with unstabilized reagents. We now report the olefination of the corresponding benzoxycarbonyl (Cbz) protected serine derived aldehyde with a number of Wittig-type reagents and under both Nozaki\(^15\) and Peterson\(^16\) conditions.

2. Results

Cbz-L-serine 4 was readily converted to the Cbz protected 4-methyl-2,6,7-trioxabicyclo[2.2.2] ortho (OBO) ester 6 and oxidized to the aldehyde 7 under conditions identical...
to those employed for the analogous Fmoc-protected compounds (Scheme 1). The Cbz-protected alcohol and aldehyde crystallize more easily than the corresponding Fmoc compound and is obtained in high ee (95%). The Boc-protected compounds were also prepared, but were obtained in reduced yields and as oils.

Reaction of the Cbz aldehyde with the stabilized ylide gave the desired olefin 9a in 77% yield from 6, with a ratio better than 95:5 E:Z geometry. Unfortunately, deprotection to give the highly unstable free β,γ-unsaturated glutamic acid was unsuccessful.

Attempts to react the aldehyde 7 with unstabilized ylides initially focussed on the simplest reagent, methylenetriphenylphosphorane, which leads to protected vinylglycine 9b. A variety of ylide generation conditions were explored (NaNH2/DMSO, NaNH2/THF, NaH/DMSO, n-BuLi/DMSO, n-BuLi/THF), with minimal success. However, KOt-Bu/Et2O, recommended as the best base to methyleneate sterically hindered ketones with triphenylmethylphosphorane, gave good yields (71%) of the alkene adduct 9b. Both the Boc and Fmoc-protected aldehydes could be olefinated under these conditions, albeit with reduced yields (54% and 6%, respectively).

Table 1. Olefination reactions with protected aldehyde 7

<table>
<thead>
<tr>
<th>Entry</th>
<th>Reagent</th>
<th>Product</th>
<th>E:Z</th>
<th>Yield (%)</th>
<th>ee (%)</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>Ph3P=CHCO2CH3</td>
<td>9a–CH=CHCO2CH3</td>
<td>&gt;95:&lt;5</td>
<td>77b</td>
<td>&gt;95</td>
</tr>
<tr>
<td>2</td>
<td>Ph3P–CH2Br</td>
<td>9b–CH=CH2</td>
<td>–</td>
<td>71b</td>
<td>77</td>
</tr>
<tr>
<td>3</td>
<td>AlMe3/CH2I2/Zn</td>
<td>9c–CH=CH2</td>
<td>–</td>
<td>76b</td>
<td>86</td>
</tr>
<tr>
<td>4</td>
<td>Ph3P–CH=CHBr</td>
<td>9d–CH=CH2</td>
<td>–</td>
<td>71b</td>
<td>72</td>
</tr>
<tr>
<td>5</td>
<td>Ph3P–CH=CHBr</td>
<td>9e–CH=CH=Et</td>
<td>17:83</td>
<td>64b</td>
<td>72</td>
</tr>
<tr>
<td>6</td>
<td>Ph3P–CH=CHBr</td>
<td>9f–CH=CH=CN</td>
<td>78:22</td>
<td>71b</td>
<td>–</td>
</tr>
<tr>
<td>7</td>
<td>Ph3P–CH=CH=CH2Br</td>
<td>9g–CH=CH=CH2</td>
<td>92:8</td>
<td>55b</td>
<td>–</td>
</tr>
<tr>
<td>8</td>
<td>Ph3P–CH2OCH3</td>
<td>9h–CH=CH=OCH3</td>
<td>63:37</td>
<td>35b</td>
<td>–</td>
</tr>
<tr>
<td>9</td>
<td>TMSCH2MgCl</td>
<td>11–CH=CH2</td>
<td>–</td>
<td>53b</td>
<td>&gt;95</td>
</tr>
</tbody>
</table>

a % ee determined after deprotection.
b Yields after two steps.
c Yields after three steps.
Figure 1. Assessment of enantiomeric purity of L-vinylglycine obtained after deprotection of (A) Cbz-L-Ser(CH₂TMS)–OBO ester 10 after ion exchange purification (B) Cbz-L-Gly(–CH=CH₂)–OBO ester 9b after ion exchange purification. Amino acid samples were prepared as described in Section 5.
The KOrBu/Et3O conditions were employed to generate a number of other unstabilized and semistabilized ylides, including 13C-labeled methylenetriphenylphosphorane (Table 1). Reaction with the Cbz-protected aldehyde produced the alkenes 9e–h in 35–71% yield from 6, with the double bond geometry corresponding to that expected for the nature of the reagent: unstabilized ylides adding with cis stereoselectivity (entry 5) while semistabilized reagents produced predominantly trans isomers (entries 6–8). In most cases the two isomers were readily separable by flash chromatography with cis possessing a lower Rf than the trans isomer.

Methylenation of the Cbz protected serine aldehyde 7 under Nozaki conditions (AlMe3/CH2I2/Zn)15 gave protected vinylglycine 9b in both good yield (76%) and ee (86%) as determined by derivatization and subsequent HPLC analysis described previously.11 Formation of the terminal alkene 9b under Peterson olefination conditions was also investigated. The β-hydroxyalkylsilane 10 was generated by Grignard reaction of trimethylsilylmethylene magnesium chloride with serine aldeyde 7 and proceeded in quantitative yield with threo stereosechemistry as previously reported.11,13c Base mediated elimination failed to afford the desired olefin 9b giving instead oxazolidinone 12 as a single diastereomer in high yield regardless of base used, temperature or length of reaction. On the other hand, acid promoted elimination and simultaneous deprotection of β-hydroxyalkylsilane 10 gave vinylglycine 11i (R=−R′=−H) in good yield (74%) with little epimerization (>95% ee) providing a route to isotopically labeled vinylglycine by incorporating the label in trimethylsilylmethylene magnesium chloride.

2.1. Deprotection

Removal of the protecting groups from 9b with TMSI, under conditions successfully used to deprotect Fmoc/OBO ester protected serine11 (neat TMSI, 80°C, 20 h, aqueous NaOH workup, cation exchange column), led to nearly quantitative conversion to α-aminobutyric acid. This was identified by comparison to authentic material by TLC, HPLC and 1H and 13C NMR. The α-aminobutyric acid was racemic as determined by both chiral HPLC analysis of the derivatized amino acid and by optical rotation. A variety of modified conditions were explored, including fewer equivalents of TMSI, dilution with CH2Cl2, reduced reaction temperature, and in situ generated TMSI. The optimum conditions (7 equiv. of TMSI diluted in CH2Cl2 at room temperature) produced the desired vinylglycine 11j (R=−R′=−H), but still resulted in 10% α-aminobutyric acid contamination. Derivatization with p-phthalaldehyde and N-i-But-L-cysteine followed by HPLC analysis showed that the α-aminobutyric acid was racemic, but the vinylglycine possessed approximately 70% ee (Fig. 1).21 Acid hydrolysis with refluxing 6N HCl has previously been used to deprotect Cbz protected methyl7a or isopropyl esters22 of vinylglycine in good yield. When the same conditions were applied to Cbz-vinylglycine-OBO ester 9b the free amino acid 11j (R=−R′=−H) was obtained in 72% yield with 71% ee when derived through Wittig olefination and 76% yield with 86% ee via Nozaki olefination. Unfortunately, recrystallization of the protected intermediate 9b does not result in enrichment of optical purity, in contrast to the resolution achieved by recrystallizing partially racemic Cbz-vinylglycine benzyl ester.7h A similar optical purity was observed with the deprotected substituted γ-ethylvinylglycine.

Since Cbz cleavage is quantitative, purification on an anion exchange column instead of a cation exchange column allows for the easy separation of any non-hydrolyzed ester. The anion exchange column also has the advantage that the only exposure of the deprotected vinylglycine to base is during the very brief neutralization while the sample is loaded onto the column.

3. Discussion

The partial racemization observed with unstabilized ylides occurs during carbonyl addition, as the aldehyde is known to be enantiomerically pure and the deprotection and derivatization reactions have been shown to cause minimal racemization.11

Nevertheless, this procedure has certain advantages over use of the Garner aldehyde, in that no oxidation step is required following alkene formation. The side chain must only be resistant to acid hydrolysis. It should be noted that the Garner aldehyde does not always lead to enantiomerically pure products; in fact with methylenetriphenylphosphorane generated by n-BuLi/THF, the alkene is obtained in 27% yield and 69% ee,23 with KH/benzene, racemic material is obtained.22 It is unclear why the propyltriphenylphosphorane derived ylide gives a product with 70% ee when reacted with the Cbz/OBO ester protected aldehyde, while giving a product with a reported >95% ee when reacted with the Garner aldehyde. Nozaki conditions give the protected vinylglycine in good yield but partial racemization is observed.

Attempts at base mediated Peterson olefination consistently resulted in formation oxazolidinone 12 in high yield regardless of base used. This may be explained by the requirement of the β-hydroxysilane to adopt the syn confirmation which is not easily achieved due to steric considerations whereas oxazolidinone formation does not suffer from this constraint. This is in contrast to recently reported synthesis of (S)-2-amino-(Z)-3,5-hexadecanoic acid in which conjugation is installed through base-mediated elimination of the Peterson olefination addition product.19 In this case the diene system does not suffer from this limitation.

The production of α-aminobutyric acid during attempts at deprotection of Cbz-vinylglycine-OBO ester with TMSI appears to be due to the presence of trace amounts of HL, which may add to the isomerized α,β-unsaturated alkene bond following OBO ester ring-opening. However, the use of 6N HCl to remove both the OBO and Cbz protecting groups proved to be very successful in giving excellent yield of vinylglycines.
4. Conclusions

We have demonstrated the ability of the serine derived aldehyde OBO ester synthon to provide entry to another class of amino acids, the vinylglycines, through olefination with a variety of reagents. Partial racemization (10–15%) with Wittig-type reagents occurs during the alkene formation, but this disadvantage is balanced by the possibilities of being able to readily synthesize a variety of unusual highly functionalized molecules, including isotopically labeled compounds that are of immense interest in mechanistic enzymology. With the Peterson olefination conditions, vinylglycine can be obtained in excellent yield and optical purity.

5. Experimental

5.1. General methods

Cbz-l-Serine was purchased from Advanced Chemtech and most other reagents from Aldrich Chemical Company and were used without further purification with the following exceptions. Zn dust was washed several times with 5% hydrochloric acid, washed with copious amounts of water, followed by methanol, then ether and dried under high vacuum. CH2Cl2, DMSO, and DIPEA were distilled from CaH2; THF and Et2O from Na/benzophenone. Reactions were carried out under Ar in glassware dried overnight at 120°C or flame dried before use.

NMR spectra were recorded in CDC13 (referenced to TMS at 0.00 ppm for 1H, to CDC13 at 77.00 ppm for 13C) or D2O (referenced to 2,2,3,3-d4-3-(trimethylsilyl)propionic acid at 0.00 ppm for both 1H and 13C) on a Bruker AC-200, AM-250 or AM-300 spectrometer. CDCl3 used for NMR measurements, if not stated otherwise, was obtained on a Kratos MALDI 3 matrix assisted laser desorption time of flight mass spectrometer. Samples were carried out by Gaston Boulay at the Universite´ de Temp apparatus in an open capillary tube and are uncorrected.

5.1.1. 3-Methyl-3-(toluenesulfonyloxymethyl)oxetane, oxetane tosylate, 3. A dry, 1 L round-bottomed flask was charged with toluene-4-sulfonyl chloride (57.20 g, 0.3 mol) to which pyridine (250 mL) was added whilst stirring under nitrogen. The reaction flask was placed inside a container to which an ice/water mixture could be added in the event that the reaction became too exothermic. 3-Methyl-3-oxetanemethanol (20.4 g, 0.2 mol) was then added slowly and the mixture stirred for 1.5 h. The mixture was then slowly added to a vigorously stirring mixture of de-ionized water (700 mL) and crushed ice (700 g) in a 2 L Erlenmeyer flask and allowed to stir for an additional 0.5 h. The white precipitate was then collected on Whatman filter paper #1 and washed with cold H2O. The product was dried under high vacuum and/or P2O5 to obtain the white powder of oxetane tosylate 3 (49.11 g, 92%). Mp 49.5–51°C. TLC (3:2, Hex/EtOAc) Rp = 0.42; 1H NMR (CDC13, 250 MHz) δ 7.81 (d, J = 8.2 Hz, 2H), 7.37 (d, J = 8.2 Hz, 2H), 4.37 (m, 4H), 4.11 (s, 2H), 2.46 (s, 3H), 1.31 (s, 3H); 13C NMR (CDCl3, 63 MHz) δ 145.1, 132.8, 129.9, 127.9, 78.9, 74.2, 39.3, 21.6, 20.6. HRMS (FAB) calcld for (M+H+) C17H13O4S 256.0769, found 256.0774. Anal. Calcld for C17H13O4S: C, 56.23; H, 3.88. Found: C, 56.33; H, 3.64.

5.1.2. N-(Benzyloxy carbonyl)-l-serine-3-methyl-3-hydroxy methyl-oxetane ester, Cbz-l-Ser-oxetane ester, 5. Cbz-l-Ser 4 (11.36 g, 0.047 mol) and Cs2CO3 (9.19 g, 0.028 mol) were combined and dissolved in H2O (100 mL). The water was then removed in vacuo and the resulting oil was lyophilized for 12 h to give a white foam. To this foam was added oxetane tosylate (12.65 g, 0.049 mol) and Nal (1.41 g, 9.8 mmol) which was then taken up in DMF (400 mL) and allowed to stir under Ar for 48 h. The DMF was then removed in vacuo and the resulting solid dissolved in EtOAc (600 mL) and H2O (200 mL) and extracted with 10% NaHCO3 (2×100 mL), saturated NaCl (100 mL) and dried over MgSO4. The solvent was removed under reduced pressure to yield a yellow oil which was recrystallized from ethyl acetate and hexanes to yield colourless rod-like crystals in 78% yield (11.85 g). Mp 70–70.5°C; [a]D20 = +76.1 (c = 0.5, H2O); TLC (Solvent A), Rp = 0.34; 1H NMR (CDCl3, 250 MHz) δ 7.35 (s, 5H), 5.89 (d, J = 7.4 Hz, 1H), 5.12 (s, 2H), 4.55–4.39 (m, 6H), 4.14–4.06 (m, 1H), 4.11 (d, J = 11.2 Hz, 1H), 3.92–3.78 (m, 1H), 3.12 (t, J = 6.0 Hz, 1H), 1.28 (s, 3H); 13C NMR (CDCl3, 62.9 MHz) δ 170.6, 156.2, 131.6, 128.5, 128.2, 128.1, 79.4, 69.0, 67.1, 63.3, 56.4, 39.6, 20.7; IR (cast from CHCl3) 3371 (br m), 3064 (vw), 3034 (vw), 2956 (m), 2879 (m), 1723 (s), 1525 (m), 1457 (w), 1397 (m), 1339 (m), 1214 (m), 1195 (m), 1061 (m), 978 (w), 834 (w), 744 (w), 699 (w) cm–1; MS (CI, CH4) m/z 324 (MH+, 100), 306 (MH+–18, 76), 293 (MH+–31, 65), 280 (MH+–44, 29); HRMS (CI, CH4) Caled for C16H12O4N: 324.1447. Found: 324.1454 (MH+). Anal. Caled for C16H12O4N: C, 59.43; H, 6.55; N, 4.33. Found: C, 59.51; H, 6.57; N, 4.36.

5.1.3. 1-[N-(Benzyloxy carbonyl)-15]-1-amino-2-ethanol]-4-methyl-2,6,7-trioxo-bicyclo[2.2.2]octane, Cbz-l-Ser-OBO ester, 6. Cbz-Ser oxetane ester 5 (15.0 g, EtOAc/hex; B, 3:1 EtOAc/hex; C, 1:1:1.1 H2O/EtOAc/n-BuOH/MeOH.
5.2. General procedure for Wittig addition to the protected aldehyde

The ylide was prepared by suspending potassium tert-butoxide (2.1 equiv.) in freshly distilled Et2O (2–4 mL) under N2. The phosphate salt or phosphate oxide (2.3 equiv.) was then added, and the resulting brightly colored suspension was refluxed for 15 min. The Et2O was then evaporated with a stream of N2 until a thick slurry was obtained, at which point a solution of crude Cbz-Ser(ald)-OBO ester 7 (1 equiv., 0.8–1.6 mmol assuming 100% yield of the aldehyde from the oxidation) in freshly distilled THF (2–3 mL) was added. After 5–10 min at 50°C, the reaction was poured into H2O (20 mL) and CH2Cl2 (20 mL). The layers were separated, the H2O was washed with CH2Cl2 (50 mL), and the organic fractions were then combined, washed with H2O (1×100 mL), dried (MgSO4), and evaporated to dryness. The residue was purified by flash column chromatography (1:1 EtOAc/hexane, loaded in CH2Cl2).

5.2.1. Wittig addition of Ph3P=CH–CO2CH3 to Cbz-l-Ser(ald)-OBO ester 7: 1-[methyl-N-(benzoyloxy carbonyl)-(1S)-1-amino-2-oxoethyl]-4-methyl-2,6,7-trioxabicyclo[2.2.2]octane, Cbz-l-Ser(ald)-OBO ester 7 (0.099 g, 0.31 mmol assuming 100% yield in the oxidation), Ph3P=CH–CO2CH3 (0.120 g, 0.359 mmol, 1.2 equiv.), and dry CH2Cl2 (6 mL) were added to a flask and stirred at room temperature. After 6 h, Et3N (1.29 mL, 9.25 mmol) was added and the solution stirred for 30 min at 0°C. Ice-cold CH2Cl2 (250 mL) was added and the resulting cloudy mixture was stirred for 1.5 h at −78°C. DIPEA (24.27 mL, 0.14 mol) was added and the solution stirred for 30 min at −78°C and 10 min at 0°C. Ice-cold CH2Cl2 (250 mL) was added and the solution was washed with ice-cold 3% NH4Cl (250 mL), 10% NaHCO3 (100 mL), and saturated NaCl (250 mL), dried (MgSO4), and evaporated to dryness. The residue was purified by flash column chromatography (1:1 EtOAc/hexane, yielded 0.089 g of white solid (77% from 6), with a >95%:<5% E/Z ratio (as determined by NMR integration of the alkenic protons). The oil crystallized upon standing. Mp 106–108°C; [α]D20 = −33.2 (c=1.16, EtOAc); TLC (Solvent B) Rf = 0.37 (Et3); 1H NMR (CDCl3, 200 MHz) δ 7.35 (s, 5H), 6.99 (dd, J = 15.8, 5.0 Hz, 1H), 6.01 (dd, J = 15.8, 1.6 Hz, 1H), 5.20 (d, J = 9.3 Hz, 1H), 5.12 (s, 2H), 4.57 (ddd, J = 8.4, 4.3, 1.4 Hz, 1H), 3.89 (s, 6H), 3.72 (s, 3H), 0.79 (s, 3H); 13C NMR (CDCl3, 50.3 MHz) δ 166.4, 155.8, 143.2, 136.2, 128.4, 128.1, 122.4, 107.6, 72.8, 67.0, 51.5, 30.7, 14.1; IR (Nujol mull) 3347 (br w), 3063 (vw), 2933 (vw), 2949 (w), 2883 (w), 2712 (s), 1660 (w), 1568 (vw), 1518 (m), 1456 (w), 1436 (w), 1309 (m), 1275 (m), 1247 (m), 1196 (m), 1172 (w), 1049 (s), 1013 (m), 987 (w), 862 (w), 750 (w), 699 (w) cm−1; MS (EI, 70 eV) Calcd for C19H23O7N: 377.1474. Found: 377.1474 ± 0.0011 (M+); Anal. Calcd for C16H20O6N: C, 59.81; H, 6.12; N, 4.33. Found: C, 59.72; H, 6.12; N, 4.33.

5.2.2. Wittig addition of Ph3P=CH2 to Cbz-l-Ser(ald)-OBO ester 4: 1-[N-(benzoyloxy carbonyl)-(1S)-1-amino-2-propenyl]-4-methyl-2,6,7-trioxabicyclo[2.2.2]octane, Cbz-l-Gly-(CH=CH2)−OBO ester 4b. Crude Cbz-Ser(ald)-OBO ester 7 (0.524 g, 1.63 mmol assuming 100% yield of
the aldehyde from the oxidation) was reacted with the yellow ylide generated from MeP^+PhBr^- according to the general procedure. Purification gave 0.368 g (71% from 6) of a thick oil ([α]D20 = −61.9 (c = 1.12, EtOAc); 75% ee by HPLC analysis). The oil could be crystallized from Et2O/hexane to give colourless crystals: mp 73–74°C; [α]D20 = −68.5 (c = 1.12, EtOAc); TLC (Solvent A) Rf 0.48; 1H NMR (CDCl3, 250 MHz) δ 7.34–7.25 (m, 5H), 5.92 (dd, J = 17.2, 10.5, 5.5 Hz, 1H), 5.31–5.06 (m, 3H), 5.12 (s, 2H), 4.43 (br t, J = 6.9 Hz, 1H), 3.89 (s, 6H), 0.78 (s, 3H), 0.76 (d, J = 7.5 Hz, 3H), 0.60 (s, J = 9.6, 17.4 Hz, 1H), 0.52 (s, J = 9.1 Hz); 13C NMR (CDCl3, 75 MHz) δ 157.2, 136.7, 128.6, 128.5, 128.2, 108.9, 72.8, 67.1, 60.5, 59.1, 30.7, 21.5, 14.3, −0.8; IR (cast from CHCl3) 3389 (br w), 3064 (vw), 3033 (vw), 2944 (w), 2881 (m), 1719 (s br), 1717 (s), 1312 (s), 1170 (s), 1051 (s), 994 (s), cm⁻¹. Anal. Calcd for C29H31NO5Si: C, 59.1; H, 6.9; N, 0.48; 1H NMR (CDCl3, 250 MHz) δ 7.40–7.26 (m, 5H), 5.32 (d, J = 10.3 Hz, 1H), 4.31 (dd, J = 4.8, 9.7 Hz, 1H), 3.89 (s, 6H), 3.68 (d, J = 10.3 Hz, 1H), 0.85 (dd, J = 9.6, 14.7 Hz, 1H), 0.78 (s, 3H), 0.60 (dd, J = 4.7, 14.7 Hz, 1H), 0.02 (s, 9H); 13C NMR (CDCl3, 75 MHz) δ 157.2, 136.7, 128.6, 128.5, 128.2, 108.9, 72.8, 67.1, 60.5, 59.1, 30.7, 21.5, 14.3, −0.8; IR (cast from CHCl3) 3389 (br w), 3064 (vw), 3033 (vw), 2944 (w), 2881 (m), 1719 (s br), 1510 (s), 1313 (s), 1170 (s), 1051 (s), 994 (s), cm⁻¹. Anal. Calcd for C29H31NO5Si: C, 58.65; H, 7.63; N, 0.48; 1H NMR (D2O, 250 MHz) δ 4.27 (d, J = 7.6 Hz, 1H), 1.90 (quintet, J = 7.1 Hz, 2H), 0.98 (t, J = 7.5 Hz, 3H); 13C NMR (D2O, 62.7 MHz) δ 177.7, 58.7, 26.5, 11.3; MS (ESI) m/z 104 (M+H+)². Derivatisation with o-phthalaldehyde and N-isobutyl-l-cysteine, and analysis by HPLC, with comparison to commercial α-aminoacibutyric acid and vinylglycine standards, indicated a 50:50 ratio of l- and d-isomers (Waters 125, 8×100 mm µ-Bondapak C18 Radial-Pak™ cartridge column, 1 mL/min; 100% 30 mM sodium acetate buffer, pH 6.5; linear gradient over 35 min to 50:50 buffer/MeOH; detection at 338 nm; diastereomers formed by l-α-aminoacibutyric acid at 28.9 min, by d-α-aminoacibutyric acid at 31.1 min, by l-vinylglycine at 26.6 min, and by d-vinylglycine at 28.5 min). The reaction time, temperature, source of TMSI and equivalents of TMSI were all varied in attempts to produce the desired vinylglycine. Ratios of α-aminoacibutyric acid and vinylglycine were determined by both 1H NMR and HPLC, as outlined above. For monitoring the reactions over time, an aliquot (5–50 µL) was removed, added to 0.5N NaOH (100–300 µL), and extracted with Et2O (1 mL). A portion of the aqueous fraction (10–40 µL) was then derivatized and analyzed as outlined above. For the in situ generation of TMSI, NaI (10 equiv.) and the alkene were dissolved in CH3CN, and TMSI/Cl (10 equiv.) was added.

(b) Acid hydrolysis of 9b with 6N HCl. Cbz-l-Gly(-CH=CH2)−OBO ester 9b (0.230 g, 0.720 mmol) was treated with TMSI (1.5 mL, 10.5 mmol, 15 equiv.) at 80°C for 20 h. After cooling, Et2O (3 mL) was carefully added, followed by the dropwise addition of 0.5N NaOH (5 mL). The organic layer was removed and washed with 0.5N NaOH (2×4 mL). The aqueous fractions were combined, washed (2×5 mL, Et2O), acidified to pH<3 with 2N HCl and purified on a cation exchange column (loaded on a Bio-Rad AG 50W-X8 100–200 mesh, hydrogen form, 1×12 cm, washed with 0.01N HCl and H2O, then eluted with 5% Et3N in H2O or, alternately, 1 M NH4H2O solution). The eluate was evaporated to dryness under vacuum gave 0.0681 g (92%) of a colorless solid, which NMR and ES−MS analysis revealed to be predominantly α-aminoacibutyric acid: [α]D20 = 0.2 (c = 0.47, AcOH); 1H NMR (D2O, 250 MHz) δ 4.27 (d, J = 7.6 Hz, 1H), 1.90 (quintet, J = 7.1 Hz, 2H), 0.98 (t, J = 7.5 Hz, 3H); 13C NMR (D2O, 62.7 MHz) δ 177.7, 58.7, 26.5, 11.3; MS (ESI) m/z 104 (M+H+)². Derivatisation with o-phthalaldehyde and N-isobutyl-l-cysteine, and analysis by HPLC, with comparison to commercial α-aminoacibutyric acid and vinylglycine standards, indicated a 50:50 ratio of l- and d-isomers (Waters 125, 8×100 mm µ-Bondapak C18 Radial-Pak™ cartridge column, 1 mL/min; 100% 30 mM sodium acetate buffer, pH 6.5; linear gradient over 35 min to 50:50 buffer/MeOH; detection at 338 nm; diastereomers formed by l-α-aminoacibutyric acid at 28.9 min, by d-α-aminoacibutyric acid at 31.1 min, by l-vinylglycine at 26.6 min, and by d-vinylglycine at 28.5 min). The reaction time, temperature, source of TMSI and equivalents of TMSI were all varied in attempts to produce the desired vinylglycine. Ratios of α-aminoacibutyric acid and vinylglycine were determined by both 1H NMR and HPLC, as outlined above. For monitoring the reactions over time, an aliquot (5–50 µL) was removed, added to 0.5N NaOH (100–300 µL), and extracted with Et2O (1 mL). A portion of the aqueous fraction (10–40 µL) was then derivatized and analyzed as outlined above. For the in situ generation of TMSI, NaI (10 equiv.) and the alkene were dissolved in CH3CN, and TMSI/Cl (10 equiv.) was added.
0.0100 g (72%) of solid. Derivatisation with o-phthalaldehyde and N-isobutyl-l-cysteine, and analysis by HPLC indicated 77% ee (conditions as outlined above): mp 178–180°C (dec); TLC (Solvent C) Rf 0.46; 1H NMR (D2O, 250 MHz) δ 5.85 (d, J = 17.4, 10.1, 7.4 Hz, 1H), 5.38 (d, J = 17.2 Hz, 1H), 5.38 (d, J = 10.4 Hz, 1H), 4.27 (d, J = 7.3 Hz, 1H); 13C NMR (D2O, 50.3 MHz): δ 174.7, 132.0, 124.8, 59.1; MS (LD, sinapinic acid) m/z 102 (MH+).

(c) Acid hydrolysis of 10 with 6N HCl. Cbz-l-Ser-(CH3TMS)–OBO ester 10 (0.140 g, 0.34 mmol) was mixed with 6N HCl (10.0 mL) and refluxed for 3 h. The solution was cooled extracted twice with ether (2×5 mL), neutralized with a saturated solution of NaHCO3 (approx. 50 mL), then loaded on an anion exchange column (Bio-Rad AG 1-X4 100–200 mesh, chloride form, converted to hydroxide form by prewashing with 4N NaOH). The column was washed with H2O, and eluted with 1N AcOH, hydroxide form by prewashing with 4N NaOH). The AG 1-X4 100–200 mesh, chloride form, converted to hydroxide form by prewashing with 4N NaOH). The column was washed with H2O, and eluted with 1N AcOH, hydroxide form by prewashing with 4N NaOH. The column was washed with H2O, and eluted with 1N AcOH, hydroxide form by prewashing with 4N NaOH. The column was washed with H2O, and eluted with 1N AcOH, hydroxide form by prewashing with 4N NaOH.

5.2.6. 1-[N-(Benzyloxycarbonyl)-(1S)-1-amino-3-13C-2-propene]-4-methyl-2,6,7-trioxabicyclo[2.2.2]octane, Cbz-l-Gly-(CH=CH2)–OBO ester 9d. Crude Cbz-Ser(ald)-OBO ester 7 (0.241 g, 0.739 mmol assuming 100% yield of the aldehyde from the oxidation) was reacted with the orange–yellow ylide generated from Ph3P=CH2–CH2Br according to the general procedure. Purification gave 0.164 g (62% from 6) of a first crop of colourless plate crystals.

5.2.7. Deprotection of Cbz-l-Gly-(CH=CH2)–OBO ester 9d: γ-13C-vinylglycine 11d. Cbz-l-Gly-(CH=CH2)–OBO ester 9d (0.071 g, 0.210 mmol) was deprotected with 6N HCl (2.0 mL) and purified by anion exchange chromatography as described above to give 0.082 g (21%) of a colourless plate. Recrystallisation (H2O/acetonitrile) gave 0.028 g (81%) of a colorless powder. Recrystallisation (H2O/acetonitrile) gave 0.028 g (81%) of a colorless powder. Recrystallisation (H2O/acetonitrile) gave 0.028 g (81%) of a colorless powder. Recrystallisation (H2O/acetonitrile) gave 0.028 g (81%) of a colorless powder. Recrystallisation (H2O/acetonitrile) gave 0.028 g (81%) of a colorless powder.

5.2.8. 1-[N-(Benzyloxycarbonyl)-(1S)-1-amino-(Z)-2-pentene]-4-methyl-2,6,7-trioxabicyclo[2.2.2]octane, Cbz-l-cis-Gly-(CH=CH=Et)–OBO ester, 9e. Crude Cbz-Ser(ald)-OBO ester 7 (0.241 g, 0.739 mmol assuming 100% yield of the aldehyde from the oxidation) was reacted with the orange–yellow ylide generated from Ph3P=CH2–CH2Br according to the general procedure. Purification gave 0.164 g (62% from 6) of a first crop of colourless plate crystals.

5.2.9. Deprotection of Cbz-l-Gly-(CH=CH2)–OBO ester 9e: Z-ethylvinylglycine, 11d. Cbz-l-Gly-(CH=CH2)–OBO ester 9e (0.071 g, 0.210 mmol) was deprotected with 6N HCl (2.0 mL) and purified by anion exchange chromatography as described above to give 0.082 g (83%) of a colorless powder, with 83:17 Z/E ratio by 1H NMR. Derivatisation with o-phthalaldehyde and N-isobutyl-l-cysteine, and analysis by HPLC indicated 72.2% ee (conditions as outlined above): mp 178–181°C (dec); TLC (Solvent C) Rf 0.46; 1H NMR (D2O, 250 MHz) δ 5.92–5.72 (m, 1H), 5.36 (ddd, J = 154.9, 17.0, 0.6 Hz, 1H), 5.36 (ddd, J = 161.1, 10.2, 0.7 Hz, 1H), 4.13 (td, J = 6.5, 0.7 Hz, 1H); 13C NMR (D2O, 62.7 MHz): δ 174.7, 132.9 (d, J = 70.1 Hz, CH=CH2), 124.0 (s, superimposed on d, J = 69.8 Hz, relative intensity 180:1, CH=CH2), 59.8; MS (LD, sinapinic acid) m/z 105 (MH+) ; Anal. Calcd for C13H20N2: H, 6.91; N, 13.72. Found: H, 7.06; N, 13.55.
5.10.1. [N-(Benzoxycarbonyl)-(1S)-1-amino-3-cyano-(E)-2-propene]-4-methyl-2,6,7-trioxabicyclo[2.2.2]octane, Cbz-l-trans-Gly-(CH=CH-CN)-OBO ester, 9f. Crude Cbz-Ser(ald)-OBO ester 7 (0.507 g, 1.54 mmol assuming 100% yield of the aldehyde from the oxidation) was reacted with the yellow ylide generated from (EtO)2PO(CH2-CN) according to the general procedure. Purification gave 0.294 g (55% from 6) of the trans isomer and 0.084 g (16%) of the cis isomer as well separated compounds (71%, 78:22 E/Z overall). The trans isomer could be recrystallized from Et2O/hexane; the cis from CH2Cl2/CH2O.

trans Isomer. Mp 106–107.5°C; [α]25D = −0.7 (c=1.37, EtOAc); TLC (Solvent A) Rf 0.44; 1H NMR (CDCl3, 250 MHz) δ 7.36 (s, 5H), 6.77 (dd, J=16.4, 4.8 Hz, 1H), 5.53 (dd, J=16.3, 1.1 Hz, 1H), 5.18–5.11 (m, 3H), 4.56–4.51 (m, 1H), 3.92 (s, 0.8), 6.12 (s, 3H); 13C NMR (CDCl3, 62.9 MHz) δ 155.8, 149.7, 136.0, 128.5, 128.3, 128.1, 116.9, 107.4, 101.7; 72.9, 67.3, 56.6, 30.8, 14.1; IR (cast from CH2Cl2) 3393 (br w), 3064 (vw), 3035 (vw), 2948 (w), 2884 (w), 2226 (w), 1720 (s), 1571 (w), 1517 (m), 1332 (w), 1253 (m), 1226 (m), 1049 (s), 1018 (s), 966 (m), 736 (w), 699 (w) cm−1; MS (EI, 70 eV) m/z 344 (M+1, 100), 253 (M+1–91, 20), 237 (M+1–107, 53); HRMS (EI, 70 eV) Calcd for C18H20O5N2: 344.1372. Found: 344.1370 (M+1).

cis Isomer. Mp 161.5–162.5°C; [α]25D = +0.9 (c=0.80, EtOAc); TLC (Solvent A) Rf 0.35; 1H NMR (CDCl3, 200 MHz) δ 7.32–7.30 (m, 5H), 6.35 (br dd, J=10.7, 8.1 Hz, 1H), 5.49 (br d, J=11.2, 1.1 Hz, 1H), 5.35 (br d, J=7.3, 1.1 Hz, 1H), 5.13 (s, 2H), 4.74 (br t, J=7.9 Hz, 1H), 3.91 (s, 6H), 0.81 (s, 3H); 13C NMR (CDCl3, 50.3 MHz) δ 155.5, 144.7, 136.1, 128.4, 128.2, 115.2, 107.4, 102.1, 72.9, 67.2, 56.0, 30.7, 14.1; IR (cast from CH2Cl2) 3355 (br w), 3062 (wv), 2949 (w), 2884 (w), 2223 (w), 1722 (s), 1652 (wv), 1634 (wv), 1512 (m), 1397 (w), 1327 (m), 1256 (m), 1223 (m), 1049 (s), 1016 (s), 1003 (s), 912 (w), 883 (w), 811 (w), 759 (w), 738 (w), 699 (w) cm−1; MS (EI, 70 eV) m/z 344 (M+1, 67), 300 (M+1–44, 8), 253 (M+1–91, 18), 237 (M+1–107, 22), 224 (M+1–120, 100); HRMS (EI, 70 eV) Calcd for C18H20O5N2: 344.1372. Found: 344.1370 (M+1); Anal. Calcd for C18H20O5N2: C, 62.78; H, 5.87; N, 8.14. Found: C, 62.64; H, 6.00; N, 8.03.

5.11. 1-[N-(Benzyloxycarbonyl)-(1S)-1-amino-2-pentene]-4-methyl-2,6,7-trioxabicyclo[2.2.2]octane, Cbz-l-cis-Gly-(CH=CH-CN)=CH2)-OBO ester, 9g. Crude Cbz-Ser(ald)-OBO ester 7 (0.257 g, 0.778 mmol assuming 100% yield of the aldehyde from the oxidation) was reacted with the orange-red ylide generated from Ph3P–CH2Br according to the general procedure. Purification gave 0.152 g (55% from 6) of predominantly trans isomer (approximately 92:8 E/Z as estimated from 13C NMR) as an oil. Recrystallization attempts resulted in a clear gel which was insoluble in most solvents:

trans Isomer. Mp 112–114°C; [α]25D = +1.3 (c=0.96, EtOAc); TLC (Solvent A) Rf 0.25; 1H NMR (CDCl3, 200 MHz) δ 7.37–7.28 (m, 5H), 6.06 (d, J=6.1 Hz, 1H), 5.12 (br s, 3H), 4.87 (t, J=9.2 Hz, 1H), 4.41 (dd, J=9.1, 6.2 Hz, 1H), 3.91 (s, 6H), 3.61 (very br s, 3H), 0.79 (s, 3H); 13C NMR (CDCl3, 50.3 MHz) δ 155.7, 149.1, 136.7, 128.3,

cis Isomer.
5.2.13. Attempted base-mediated Peterson olefination of Cbz-L-Ser(CH2TMS)–OBO ester, 10. (5S)-4-(4-methyl-2,6,7-trioxabicyclo[2.2.2]oct-1-yl)-5-[(1,1,1-trimethylsilyl)-methyl]-1,3-oxazolan-2-one, 12. Cbz-L-Ser(CH2TMS)–OBO ester 10 (0.210 g, 0.52 mmol) was dissolved in dry THF (20 mL) and rapidly transferred to a flask containing KH (0.063 g, 0.55 mmol, washed with Et2O) suspended in THF (10 mL) at −10°C under Ar. One hour later a TLC indicated total conversion to 12. TLC (1:2 EtOAc/hexane), Rf = 0.41; 1H NMR (CDCl3, 250 MHz) δ 5.38 (s, 1H), 4.66 (dd, J = 3.4 Hz, 1H), 3.85 (s, 6H), 3.30 (d, J = 3.4 Hz, 1H), 1.90 (dd, J = 14.5, 7.8 Hz, 1H), 0.95 (dd, J = 14.5, 6.8 Hz, 1H), 0.76 (s, 3H), 0.02 (s, 9H); 13C NMR (CDCl3, 62.9 MHz) δ 158.9, 107.4, 76.3, 72.7, 62.8, 30.8, 24.7, 14.1, –1.1; Anal. Calcld for C18H23O6N: C, 51.80; H, 7.69; N, 4.64. Found: C, 51.80; H, 7.69; N, 4.34.

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References


20. Brückner, H.; Wittner, R.; Godel, H. J. Chromatogr. 1989, 476, 73–82. Difficulty was observed in obtaining good yields of N-i-Bu-l-Cys without racemization when the procedure reported by Brückner, Wittner and Godel was used (4–5% racemization observed). Reaction of l-Cys in 2:1 H2O/dioxane with 10 equiv. of NaOH and 5 equiv. of isobutyryl chloride for 10 min was found to give much better yields of (N-i-Bu-l-Cys)2 with minimal racemization (<0.2%). Reduction with Zn/2N HCl gave the desired N-i-Bu-l-Cys.

21. Commercial l-vinylglycine (Aldrich) gave 97.2% ee when assayed under identical conditions, indicating that the slightly basic derivatization procedure was not responsible for any significant racemization.