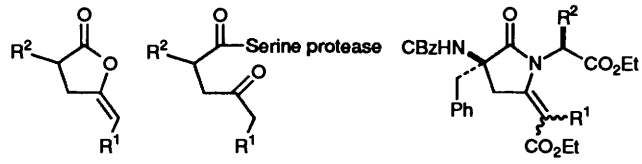
Synthesis of Phenylalanine-based Cyclic Acylated Enamino Ester Dipeptide Analogues: Inhibitors of α-Chymotrypsin. X-Ray Molecular Structure of (2'*S*,4'*R*)-4'-Benzyl-3'-benzyloxycarbonyl-5'-oxo-2'-phenyloxazolidin-4'-ylacetic Acid

Andrew D. Abell,* Mark D. Oldham and Jane M. Taylor
Department of Chemistry, University of Canterbury, Christchurch, New Zealand

Alkylation of the (*S*)-phenylalanine-derived *syn*-oxazolidinone **8** with $BrCH_2CO_2CHPh_2$ gave compound **9**, a key precursor to the β-keto ester **11** and the keto acid phosphorane **17**. Compound **17** gave the enolactone **24** on heating and the bromo enolactones **19** and **20** on treatment with bromine and triethylamine. Compounds **11**, **19**, **20** and **24** were treated with glycine ethyl ester to give the Phe-Gly dipeptide mimics **22**, **23** and **26**. The enolactone **24** also gave the Phe-Ala mimic **39** and the Phe-Gly-Gly mimic **34**.

The biostability, selectivity and potency of a peptide-based enzyme inhibitor is often enhanced by the introduction of a conformational constraint, for example a lactam ring, into the molecule.[1] Highly specific enzyme inhibitors have also been produced by introducing latent reactivity into a substrate-peptide mimic.[2] The latent reactivity is specifically released by the target enzyme to give the active inhibitor.[2] For example, halogeno enolactones **1** are simple amino acid analogues that inhibit serine proteases[2,3] by the specific release of a highly electrophilic, enzyme-bound, α-halogeno ketone **3**.[4] The related protio enolactones **2** are alternative substrate inhibitors of serine proteases.[5] Little is known about the nature of the conformational restriction imposed by the lactone rings in compounds **1** and **2** with regard to the potency of inhibition, although some active-site-modelling studies have been reported.[4c] In a preliminary communication we reported a new class of lactam-based dipeptide mimic **5** and the related system **4** which contains a latent reactive bromo enamine group.[6] These compounds represent peptide-based extensions of the protio and halogeno enolactone serine protease inhibitors, **1** and **2**, discussed above. In this paper we present two synthetic approaches to phenylalanine-based examples of these peptide mimics. The phenylalanine functionality was chosen as α-chymotrypsin is known to cleave peptides on the carboxy-group side of aromatic amino acids.[2-4]



1 R^1 = Halogen, **2** R^1 = H

3 R^1 = Halogen

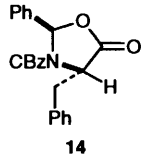
4 R^1 = Br, R^2 = H; **5** R^1 = H, R^2 = H or Me

CBz = $PhCH_2OCO$—

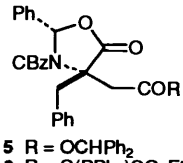
Results and Discussion

The key *syn*-oxazolidinone **8** was prepared by the methods of Seebach and Fadel[7] (step i, Scheme 1) and also Karady *et al.*[8] (step ii, Scheme 1). The former method involved the reaction of benzyl chloroformate with the Schiff base sodium salt of benzaldehyde and (*S*)-phenylalanine **6**. The second method involved treatment of (*S*)-(*N*-benzyloxy carbonyl)(CBz)-phenylalanine **7** with benzaldehyde and toluene-*p*-sulfonic acid (PTSA) with azeotropic removal of water. A ^1H NMR spectrum of the crude product mixture revealed the presence of less than 5% of the corresponding *anti*-epimer **14** in the case of step i (Scheme 1) and 50% of the *anti*-epimer **14** for step ii (Scheme 1). Silica chromatography and recrystallisation gave the pure *syn*-oxazolidinone **8** in an overall yield of 47% for method 1 and 21% for method 2. The assignment of *syn* and *anti* configurations to the oxazolidinones is discussed later.

The oxazolidinone **8** was alkylated, with >95% diastereoselectivity, using the general method pioneered by Seebach;[7,9] a tetrahydrofuran (THF) solution of the oxazolidinone **8**, at −78 °C, was treated with lithium hexamethyldisilazide (LiHMDS) followed by either $BrCH_2CO_2CHPh_2$ or $BrCH_2COC(PPh_3)CO_2Et$ (Scheme 1, steps iii and viii respectively). The crude oxazolidinones (**9** and **13**) contained less than 5%, by ^1H NMR spectroscopy, of the minor *anti*-epimers **15** and **16**, respectively. Compound **13** was obtained in 26% yield after radial chromatography. Resonances in the ^1H and ^{13}C NMR spectrum of the CBz-protected phosphorane **13**, at 23 °C, were doubled presumably due to restricted rotation about the CBz group. However, a ^1H NMR spectrum of compound **13** in $(CD_3)_2SO$ ($[^2H_6]$DMSO), at 85 °C, indicated that a single conformer was present. The crude benzhydryl oxazolidinone **9**, which was obtained quantitatively, was subsequently used without further purification. The stereochemical outcome of the alkylations is the result of self reproduction of chirality, *i.e.* the formation of the *syn*-oxazolidinones **9** and **13** proceeds with retention of configuration.

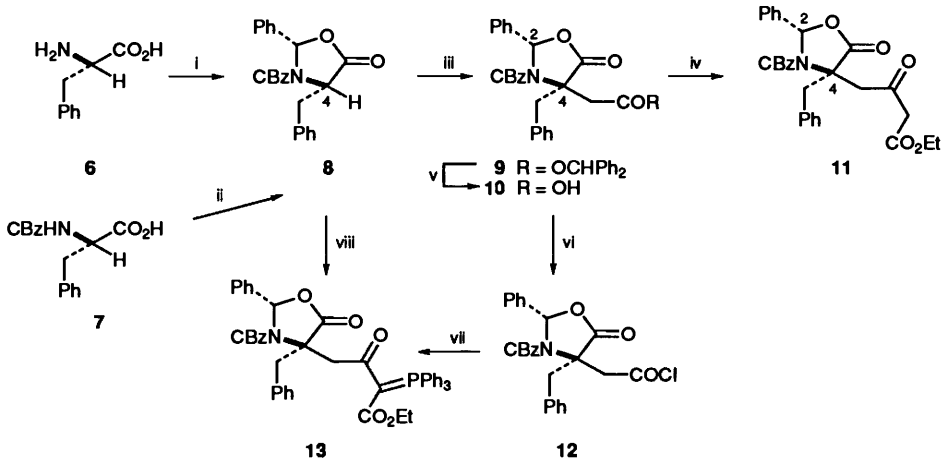


14



15 R = $OCHPh_2$
16 R = $C(PPh_3)CO_2Et$

The benzhydryl group was removed from compound **9** on treatment with trifluoroacetic acid (TFA) at 0 °C to give the acid **10** (step v, Scheme 1). The acid **10** was converted into the acid chloride **12** by use of oxalyl dichloride and a catalytic quantity of dimethylformamide (DMF). Treatment of the acid chloride **12** with 2 mol equiv. of $Ph_3P{=}CHCO_2Et$ followed by radial chromatography gave the phosphorane **13** in quantitative yield. Although requiring three additional steps, the



Scheme 1 *Reagents and conditions:* i, NaOH, PhCHO; then $PhCH_2OCOCl$, −20 to 4 °C; ii, PhCHO, PTSA, Cl_3CMe, reflux; iii, LiHMDS, THF, −78 °C; then $BrCH_2CO_2CHPh_2$; iv, THF; then $Mg(O_2CCH_{20}CO_2Et)_2$; v, TFA, CH_2Cl_2, 0 °C; vi, $(COCl)_2$, DMF, CH_2Cl_2; vii, Ph_3PCHCO_2Et (2 mol equiv.), CH_2Cl_2; viii, LiHMDS, THF, −78 °C; then $BrCH_2COC(PPh_3)CO_2Et$

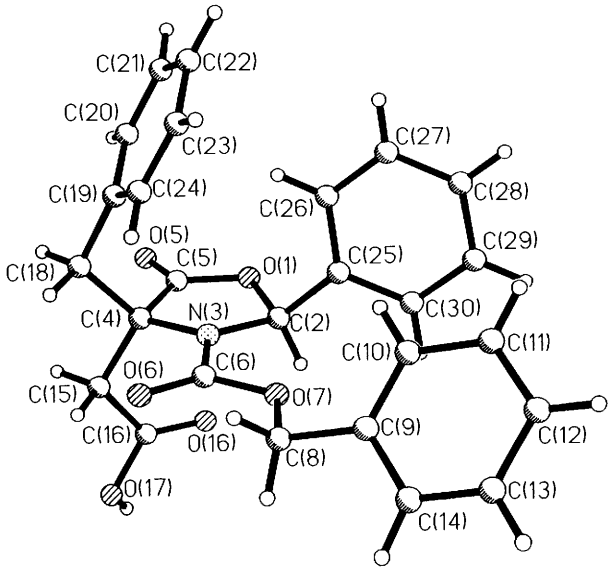


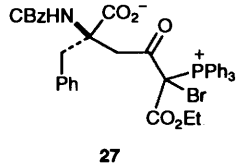
Fig. 1 X-Ray molecular structure of compound **10** with crystallographic numbering scheme

phosphorane **13** was prepared in superior yield (64%) and purity by this method, rather than *via* the direct alkylation of the oxazolidinone **8** with $BrCH_2COC(PPh_3)CO_2Et$ (step viii, Scheme 1). The acid **10** was also converted into the β-keto ester **11** in 65% yield on treatment with carbonyldiimidazole (CDI) followed by magnesium bis(ethyl malonate) (step iv, Scheme 1). Compounds **11** and **13** were key synthetic intermediates to the target peptide mimics (see later).

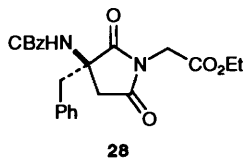
Assignment of the Oxazolidinone syn/anti *Configuration.*—The upfield position of the 4-H resonances in the major oxazolidinone isomer (assigned to structure **8**) relative to the minor isomer (assigned to structure **14**) is consistent[7,10] with the indicated *syn*/*anti* configurations. The oxazolidinone **8** also gave identical IR and ^{13}C NMR data with those previously reported.[8] However, in this initial report[8] some ambiguity exists in the reported X-ray structure and in the representation of (*S*)-phenylalanine. We have subsequently confirmed the *syn* assignment to compound **8** by an independent single-crystal X-ray structure assignment.[11] The configuration of the 4,4-disubstituted CBz-oxazolidinones **9**–**13** was consistent with the observation of a nuclear Overhauser enhancement (NOE) between 2-H and 4-CH_2CO in the β-keto ester **11** and also a single-crystal X-ray analysis of compound **10**, Fig. 1.

Synthesis of the Peptide Mimics.—Hydrolysis of the oxazolidinone ring of compound **13** with a large excess of LiOH gave the keto acid phosphorane **17** quantitatively (step i, Scheme 2). The keto acid phosphorane **17** was relatively unstable and was used subsequently without further purification. However, methylation with diazomethane, followed by radial chromatography, gave the corresponding methyl ester **18**, which was fully characterised. The acid **17** and methyl ester **18** existed as single conformers by ^1H NMR spectroscopy, unlike the precursor oxazolidinone **13** discussed earlier.

The keto acid phosphorane **17** was refluxed in THF for 6 h to give the protio enolactone **24**, which was isolated in 73% yield following radial chromatography (step vi, Scheme 2). Bromolactonisation of the keto acid phosphorane **17** with Br_2 and triethylamine gave the (*Z*)- and (*E*)-bromo enolactones **19** and **20**, respectively) in the ratio 54% *Z*:46% *E* by ^1H NMR spectroscopy. The isomers were separated by silica gel radial chromatography. The halogenolactonisation of keto acid phosphoranes is discussed in detail elsewhere;[12,13] however, a phosphonium salt of the type **27** is thought to be a reaction intermediate.

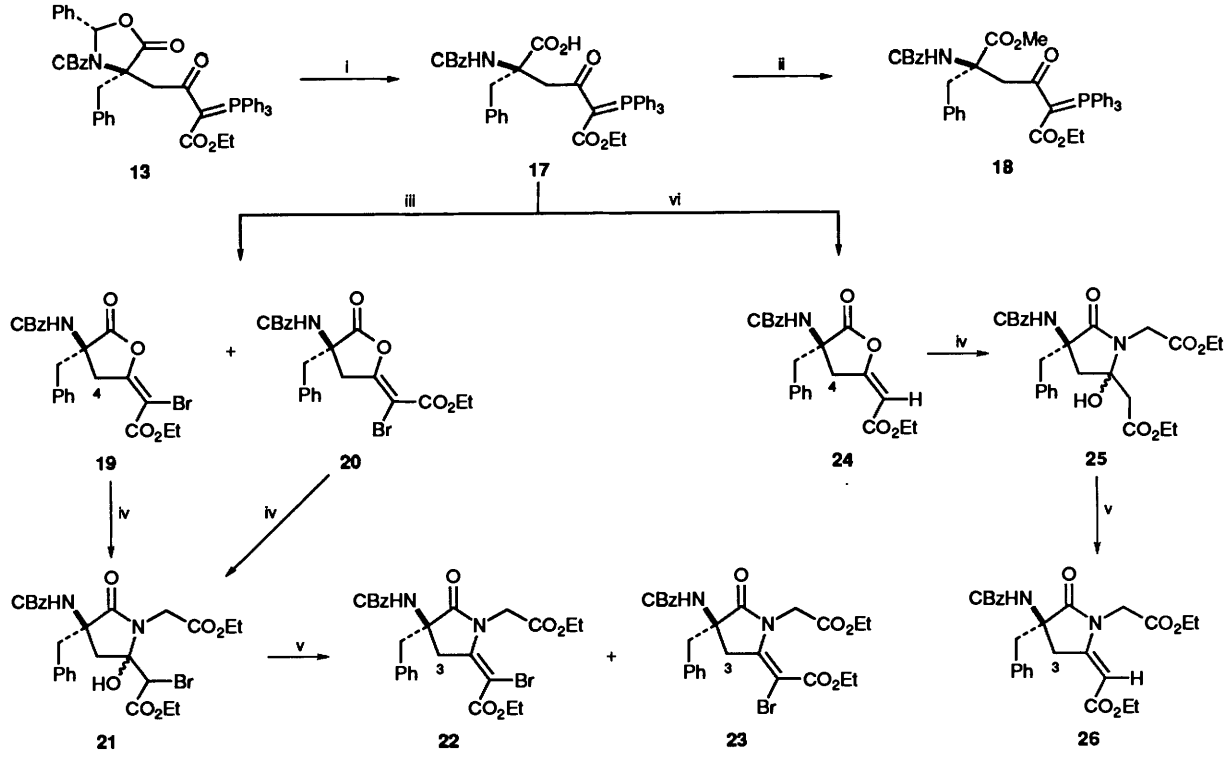


27

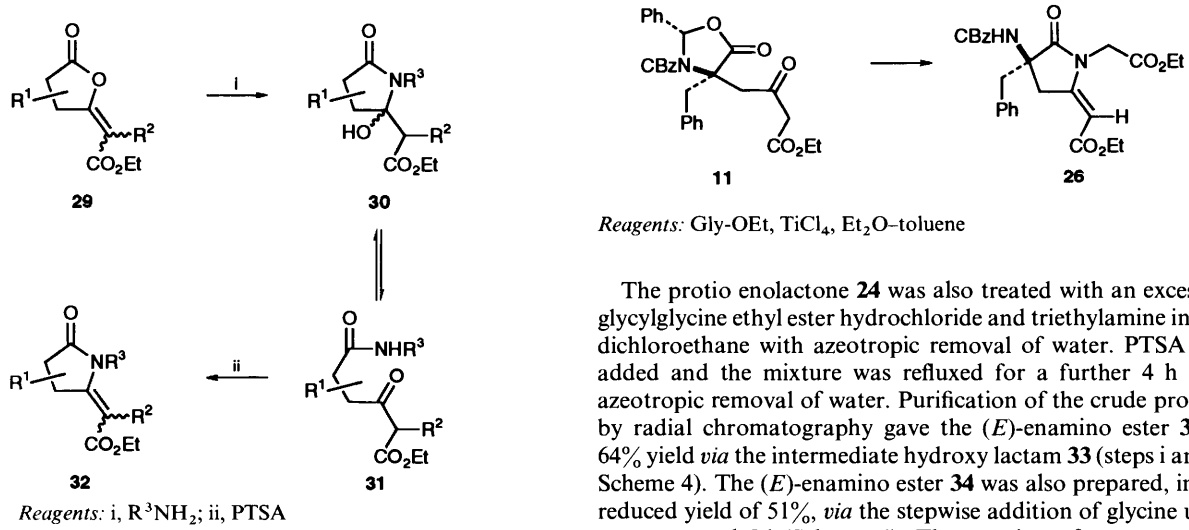


28

The protio enolactone **24** and the bromo enolactones **19** or **20** were each dissolved in dichloromethane and the solutions were stirred for 16 h at 20 °C with glycine ethyl ester hydrochloride and triethylamine to yield the corresponding hydroxy lactams **25** and **21**, respectively (Scheme 2). Compound **25** was observed, by ^1H NMR spectroscopy, to exist as a mixture of isomers in the ratio 9:1, while the bromo compound **21** existed as a complex mixture of diastereoisomers. In general, the reaction of an enolactone **29** with an amine can give either a hydroxy lactam **30** or a keto amide **31** depending on the substitution pattern of the anhydride (Scheme 3).[14] Compounds **30** and **31** give rise to the enolactam **32** on treatment with toluene-*p*-sulfonic acid



Scheme 2 *Reagents and conditions:* i, LiOH, THF, MeOH, reflux; ii, CH_2N_2, THF; iii, Br_2, Et_3N, CH_2Cl_2; iv, HCl·Gly-OEt, Et_3N, CH_2Cl_2; v, PTSA, $(CH_2)_2Cl_2$, reflux; vi, THF, reflux



Reagents: Gly-OEt, $TiCl_4$, Et_2O–toluene

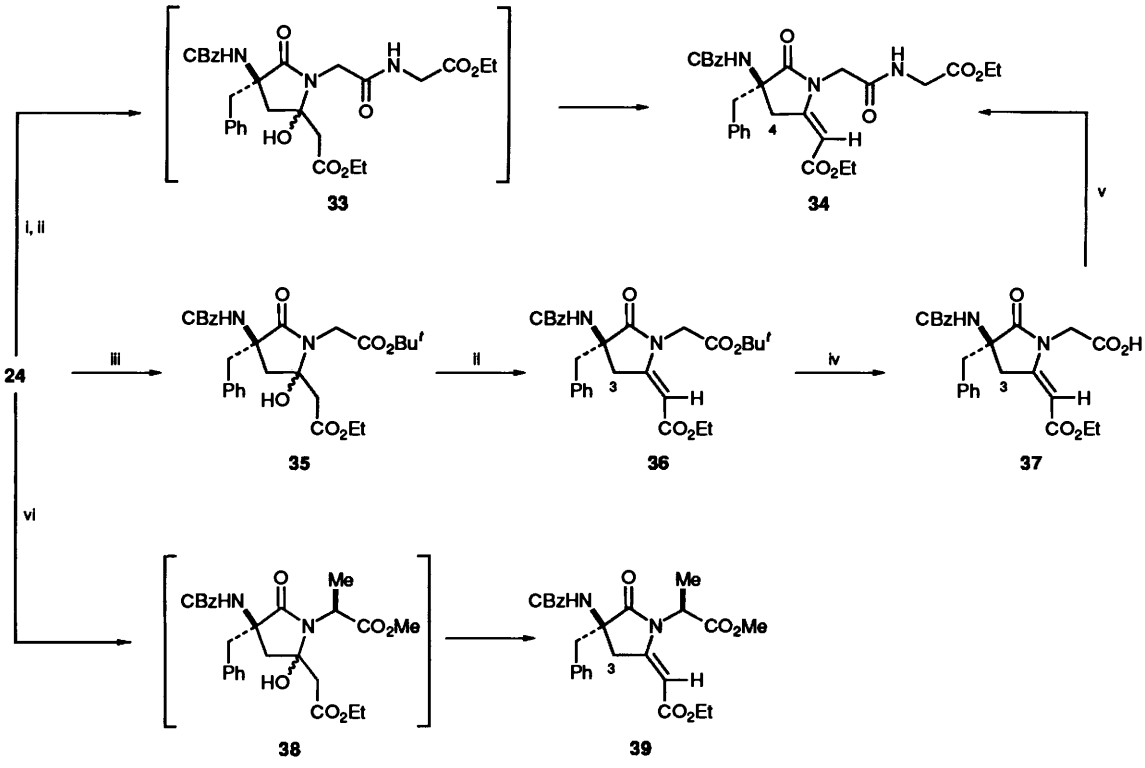
Scheme 3 *Reagents:* i, R^3NH_2; ii, PTSA

(PTSA).[14] Substituted anhydrides such as **19**, **20** or **24** tend to give the cyclic hydroxy lactam, *e.g.* **21** or **25**, rather than the corresponding acyclic keto amide intermediate.

The hydroxy lactams **25** and **21** were each dissolved in 1,2-dichloroethane, containing PTSA, and refluxed with azeotropic removal of water to give the enamino esters **26** and a mixture of geometric isomers **22** and **23**, respectively (Scheme 2). The crude enamino esters were purified by radial chromatography to give the (*E*)-enamino ester **26** in 68% yield and an inseparable mixture of the (*Z*)- and (*E*)-bromoenamino esters (**22** and **23**, 85:15 by ^{1}H NMR spectroscopy in 65% combined yield). The reaction of glycine ethyl ester with either the (*Z*)-bromo enolactone **19** or the (*E*)-bromo enolactone **20** gave hydroxy lactam **21** as a common intermediate and the same ratio of final products, compounds **22** and **23**. The imide **28** was also isolated from the crude bromo enamino ester reaction mixture in 13% yield. The enamino ester **26** was also prepared, in low yield, by a $TiCl_4$-catalysed reaction of the β-keto ester **11** with glycine ethyl ester.

The protio enolactone **24** was also treated with an excess of glycylglycine ethyl ester hydrochloride and triethylamine in 1,2-dichloroethane with azeotropic removal of water. PTSA was added and the mixture was refluxed for a further 4 h with azeotropic removal of water. Purification of the crude product by radial chromatography gave the (*E*)-enamino ester **34** in 64% yield *via* the intermediate hydroxy lactam **33** (steps i and ii, Scheme 4). The (*E*)-enamino ester **34** was also prepared, in the reduced yield of 51%, *via* the stepwise addition of glycine units to compound **24** (Scheme 4). The reaction of compound **24** with glycine *tert*-butyl ester hydrochloride and triethylamine in dichloromethane gave the corresponding hydroxy lactam **35** as a mixture of diastereoisomers in the ratio 9:1, by ^{1}H NMR spectroscopy. The hydroxy lactam **35** was dissolved in 1,2-dichloroethane containing PTSA and the solution was refluxed for 3 h, with azeotropic removal of water, to give the *tert*-butyl (*E*)-enamino ester **36**. Further PTSA was added to a solution of compound **36** in benzene, and the solution was refluxed, with azeotropic removal of water, for 3 h to give the deprotected (*E*)-enamino ester **37**. Finally, compound **37** was treated with *N*,*N*′-dicyclohexylcarbodiimide (DCC), glycine ethyl ester hydrochloride and triethylamine to give the (*E*)-enamino ester **34**, which was purified by radial chromatography.

The protio enolactone **24** was treated with an excess of (*S*)-alanine methyl ester hydrochloride and triethylamine, in 1,2-dichloroethane, with azeotropic removal of water to give, *via* intermediate hydroxy lactam **38**, the crude (*E*)-enamino ester **39** which was isolated in 78% yield following radial chromato-

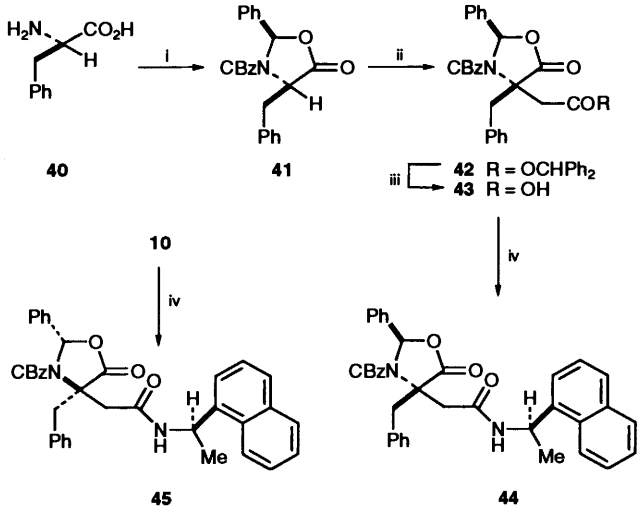


Scheme 4 *Reagents and conditions:* i, HCl·GlyGly-OEt, Et_3N, $(CH_2)_2Cl_2$, reflux; ii, PTSA, $(CH_2)_2Cl_2$, reflux; iii, HCl·Gly-OBu^t, Et_3N, CH_2Cl_2; iv, PTSA, benzene, reflux; v, HCl·Gly-OEt, Et_3N, DCC, CH_2Cl_2; vi, HCl·(*S*)-Ala-OMe, Et_3N, PTSA, $(CH_2)_2Cl_2$, reflux

graphy. The ^1H and ^{13}C NMR spectra of compound **39** were consistent with it consisting of >90% of a single isomer. Therefore, all the reactions leading to compound **39**, and by analogy compounds **22**, **23**, **26** and **34** (derived from the common precursor **10**), occur with a high degree of stereocontrol. The general procedure developed by Seebach for the preparation of α,α-dialkylated amino acids, and used here in the preparation of *syn*-oxazolidinone **9** and hence acid **10**, is reported to proceed with high diastereoselectivity.[9] For comparison, the configurational purity of acid **10** was determined by coupling to (*R*)-(+)-1-(1-naphthyl)ethylamine to give **45** (step iv, Scheme 5). Compound **44** was prepared as a reference from (*R*)-phenylalanine **40** (steps i–iv, Scheme 5). Compounds **44** and **45** gave completely different ^1H NMR spectra. There was no evidence of isomers in the ^1H NMR spectra of the crude samples of compounds **44** and **45**. Compounds **44** and **45** were subsequently purified by chromatography and fully characterised.

Assignment of E/Z *Configuration.*—The configurations of the enolactones **19**, **20** and **24** and the enamino esters **22**, **23**, **26**, **34**, **36**, **37** and **39** were assigned on the basis of ^1H NMR spectroscopy. The (*Z*)-isomer **19** was assigned on the basis of a downfield position of the 3-H_2 resonance, relative to that in the (*E*)-isomer **20**, which reflects the deshielding influence of CO_2Et.[13,15] Other characteristic differences between the ^1H NMR spectra of the (*E*)- and (*Z*)-bromo enolactones were as follows; the 3-H_2 protons appeared as a well separated AB quartet in the (*Z*)-isomer **19** and as an overlapping multiplet in the (*E*)-isomer **20**, and OCH_2Me appeared as a multiplet in the (*Z*)-isomer **19** and as a quartet in the (*E*)-isomer **20**. The ylidene carbon, C-2, resonance was downfield in the (*Z*)-isomer **19** (δ_C 159.71) relative to the (*E*)-isomer **20** (δ_C 155.25), a trend also observed[13] for related chloro and bromo enolactones. Proton–carbon heteronuclear correlation NMR experiments were used to assign the ^1H and ^{13}C spectra.

The major bromo enamino ester was assigned the *Z*

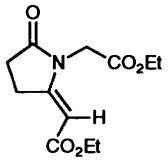


Scheme 5 *Reagents and conditions:* i, ref. 7; ii, LiHMDS, THF, −78 °C; then $BrCH_2CO_2CHPh_2$; iii, TFA, CH_2Cl_2, 0 °C; iv, (*R*)-(+)-1-(1-naphthyl)ethylamine, DCC, CH_2Cl_2, HOBt

configuration **22** due to the similarity of its ^1H NMR spectrum with that of (*Z*)-bromo enolactone **19**. The resonances for 4-H_2 appeared at similar chemical shifts to those of the (*Z*)-bromo enolactone **19**. The multiplicity of the 3-H_2 and OCH_2Me resonances of the major bromo enamino ester **22** were also the same as in (*Z*)-bromo enolactone **19**; namely, an AB quartet and multiplet, respectively. The enolactone **24** and the enamino esters **26**, **34**, **36**, **37** and **39** were assigned the *E*-configuration on the basis of the downfield positions and multiplicity (ABq) of the 3-H_2 resonances. Model studies[15] have also revealed that the cyclisation of a keto acid phosphorane of the type **17** generally gives the (*E*)- rather than the (*Z*)-protio enolactone, *e.g.* **24**. Similarly, the insertion of an amine into an enolactone

of the type **24** generally gives an enamino ester with the (*E*)-configuration, *e.g.* **26**.[14]

Preliminary results indicate that extension of the peptide sequence of the mimics results in an increase in the potency of α-chymotrypsin inhibition. For example, compound **46** is a very poor inhibitor of α-chymotrypsin and the lactams **22**/**23** (40% inhibition of α-chymotrypsin at an inhibitor concentration of 0.35 mmol dm^{-3})†,[16] and **26** (40% inhibition of α-chymotrypsin at an inhibitor concentration of 0.40 mmol dm^{-3})† are more potent inhibitors of α-chymotrypsin than the corresponding lactones **19** (35% inhibition of α-chymotrypsin at an inhibitor concentration of 0.45 mmol dm^{-3})† and **24** (25% inhibition of α-chymotrypsin at an inhibitor concentration of 0.49 mmol dm^{-3}),† respectively. Ongoing work is centred on a detailed analysis of the α-chymotrypsin inhibition and also incorporating the peptide mimics into more specific recognition peptides.



46

In conclusion, a new general route to enamino esters, involving reaction of an amino acid-derived enolactone and a second amino acid, has been developed and used to synthesise a new class of conformationally restricted dipeptide mimic. Examples of this new class, compounds **22** and **23**, possess a latent reactive bromo enamine functionality. The enolactones were prepared by the bromo enolactonisation of a keto acid phosphorane. An alternative route to the enamino esters by reaction of a β-keto ester with an amino acid was also developed.

Experimental

General.—Mps were taken using a Reichert hot-stage microscope and are uncorrected. Optical rotations were measured on a JASCO J-20C recording spectropolarimeter, and $[\alpha]_D$-values are given in units of 10^{-1} deg cm^2 g^{-1}. IR spectra were recorded on either a Pye Unicam SP3-300 or a Perkin-Elmer 1600 Series FTIR spectrometer. ^1H and ^{13}C NMR spectra were recorded on a Varian CFT300 spectrometer for samples in $CDCl_3$ solution (unless otherwise stated) with Me_4Si as internal standard. *J* Values are given in Hz. NMR locants for **39** refer to the systematic name given, and do not necessarily correspond with the text in the Results and Discussion section. Mass spectra were obtained using a Kratos MS80RFA spectrometer. Radial chromatography was performed on a chromatotron (Harrison and Harrison) using Merck type 60 PF_{254} silica gel. Light petroleum refers to the fraction of distillation range 60–70 °C.

(2S,4S)-*Benzyl*4-*Benzyl*-5-*oxo*-2-*phenyloxazolidine*-3-*carboxylate* **8**.—*Method A*. The Schiff base salt[7] (0.121 mol) of (*S*)-phenylalanine and benzaldehyde, as a solution in CH_2Cl_2 (500 cm^3), was cooled to −20 °C and benzyl chloroformate (17.0 cm^3, 0.121 mol) was added. The mixture was stirred at −20 °C for 12 h and then at 4 °C for 3 days. The solvent was evaporated off and the residue was partitioned between ethyl acetate (500 cm^3) and 5% aq. $NaHCO_3$ (500 cm^3). The organic layer was extracted, washed successively with 5% aq. $KHSO_4$ (500 cm^3) and water (500 cm^3), dried (Na_2SO_4), and evaporated to yield an oil which contained, by ^1H NMR, 95% *syn*-oxazolidinone **8** and 5% *anti*-oxazolidinone **14**. Purification by silica column chromatography and elution with light petroleum–ethyl acetate (4:1) gave the *syn*-oxazolidinone **8** (22.06 g, 47%), mp 124–126 °C (from ethyl acetate–light petroleum) (lit.,[8] 109–112 °C); δ_H 3.19–3.43 (2 H, br m, 4-CH_2Ph), 4.66 (1 H, dd, *J* 4.0, 5.9, 4-H), 5.05 and 5.16 (2 H, ABq, *J* 12.1, OCH_2Ph), 6.45 (1 H, br s, 2-H) and 7.06–7.33 (15 H, m, ArH); δ_C 36.38, 58.13, 67.76, 89.10, 126.55, 127.10, 127.98, 128.11, 128.27, 128.39, 128.58, 129.16, 130.15, 135.13, 136.12, 153.75 and 170.90. Selected ^1H NMR data for the *anti*-oxazolidinone **14**; δ_H 3.11 (2 H, m, CH_2Ph) and 4.71 (1 H, m, 4-H).

Method B.[8] (*S*)-CBz-phenylalanine **7** (10.0 g, 0.033 mol), benzaldehyde (6.8 cm^3, 0.067 mol, 2 mol equiv.) and PTSA (6.36 g, 0.033 mol, 1 mol equiv.) were dissolved in 1,1,1-trichloroethane (135 cm^3) and the solution was refluxed, with azeotropic removal of water, for 18 h to give the crude *syn*- and *anti*-oxazolidinones (**8** and **14**, respectively) in the ratio 1:1 by ^1H NMR spectroscopy. The *syn*-oxazolidinone **8** was purified as above (21%); mp and ^1H NMR as given above.

(2′S,4′R)-4′-*Benzyl*-3′-*benzyloxycarbonyl*-5′-*oxo*-2′-*phenyloxazolidin*-4′-*ylacetic Acid* **10**.—The oxazolidinone **8** (7.85 g, 0.020 mol, 1 mol equiv.) was dissolved in THF (200 cm^3) and the solution was cooled to −78 °C. LiHMDS (22.3 cm^3 of 1 mol dm^{-3} solution in THF; 0.022 mol, 1.1 mol equiv.) was added and the solution was stirred at −78 °C for 7 min. $BrCH_2CO_2CHPh_2$ (6.43 g, 0.0211 mol, 1.04 mol equiv.) was added and the resulting yellow solution was stirred at −78 °C for 2 h and was then allowed to warm to 20 °C during 16 h. The THF was evaporated off and the residue was partitioned between saturated aq. NH_4Cl (100 cm^3) and diethyl ether (100 cm^3). The aqueous layer was separated, and extracted with diethyl ether (2 × 100 cm^3). The combined extracts were dried (Na_2SO_4) and evaporated to give the crude oxazolidinone **9** as a yellow oil (12.3 g, quant), which was used in subsequent steps without further purification; δ_H 3.19 and 3.89 (2 H, ABq, *J* 17.4, C$H_2CO_2CHPh_2$), 3.25 and 3.56 (2 H, ABq, *J* 13.2, 4-CH_2Ph), 4.66 and 5.02 (2 H, ABq, *J* 12.7, OCH_2Ph), 5.95 (1 H, s, 2-H, 5.99 (2 H, d, *J* 7.3, ArH), 6.61 (2 H, d, *J* 7.3, ArH), 6.91 (1 H, s, CHPh$_2$), 6.93 (2 H, m, ArH) and 7.07–7.36 (19 H, m, ArH).

The benzhydryloxazolidinone **9** (12.40 g, 0.020 mol) was dissolved in CH_2Cl_2 (500 cm^3) and the solution was cooled to 0 °C. TFA (31 cm^3, 0.406 mol, 20 mol equiv.) was added and the solution was stirred at 0 °C for 2 h, then was diluted to 1 dm^3 with CH_2Cl_2 and washed with water (3 × 1 dm^3). The organic layer was dried ($MgSO_4$) and the solvent was evaporated off to yield the *acid* **10** as a yellow oil, which was crystallised from ethyl acetate–light petroleum (5.75 g, 64%), mp 181–185 °C (Found: C, 69.4; H, 5.4; N, 3.1. $C_{26}H_{23}NO_6 \cdot 1/4H_2O$ requires C, 69.34; H, 5.26; N, 3.11%); ν_{max}(KBr)/cm^{-1} 3415, 1794, 1738 and 1674; δ_H 3.13 and 3.87 (2 H, ABq, *J* 18.1, CH_2CO_2H), 3.25 and 3.57 (2 H, ABq, *J* 13.5, 4-CH_2Ph), 4.82 and 5.11 (2 H, ABq, *J* 12.2, OCH_2Ph), 6.13 (2 H, d, *J* 7.3, ArH), 6.30 (1 H, s, 2-H), 6.68 (2 H, d, *J* 7.4, ArH) and 6.96–7.41 (11 H, m, ArH); δ_C 38.85, 41.85, 65.00, 67.45, 90.56, 127.71, 127.96, 128.21, 129.13, 129.35, 130.82, 134.65, 136.16, 135.40, 152.24, 172.72 and 174.75.

(2′S,4′R)-(−)-*Ethyl* 4-(4′-*Benzyl*-3′-*benzyloxycarbonyl*-5′-*oxo*-2′-*phenyloxazolidin*-4′-*yl*)-3-*oxobutanoate* **11**.—CDI (175 mg, 1.08 mmol, 1.2 mol equiv.) was added to a solution of acid **10** (400 mg, 0.90 mmol) in THF (40 cm^3). After stirring of the mixture at 20 °C for 2 h, freshly prepared magnesium bis(ethyl malonate)[17] (257 mg, 0.90 mmol, 1 mol equiv.) was added and the mixture was stirred at 20 °C for 19 h. The mixture was concentrated to 5 cm^3, diluted with ethyl acetate (35 cm^3) and washed successively with water (40 cm^3), 5% aq. $KHSO_4$ (40

† Preliminary α-chymotrypsin inhibitory activities were measured using a microtitre plate-based colorimetric assay; see ref. 16.

cm^3), 5% aq. $NaHCO_3$ (40 cm^3) and 10% aq. NaCl (40 cm^3). The organic layer was dried (Na_2SO_4) and the solvent was evaporated off. Purification by radial chromatography using a 2 mm silica gel chromatotron plate, and elution with light petroleum–ethyl acetate (3:1) and crystallisation from ethyl acetate–light petroleum gave *compound* **11** (300 mg, 65%), mp 118–121 °C (Found: C, 69.7; H, 5.6; N, 2.65. $C_{30}H_{29}NO_7$ requires C, 69.89; H, 5.67; N, 2.72%); $[\alpha]_D^{20}$ −1 (*c* 15.5, CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 1791 and 1714; δ_H 1.31 (3 H, t, *J* 7.1, Me), 3.20 and 3.52 (2 H, ABq, *J* 13.2, 4-CH_2Ph), 3.28 and 4.10 (2 H, ABq, *J* 18.8, 4-CH_2CO), 3.46 (2 H, s, $COCH_2CO$), 4.23 (2 H, q, *J* 7.1, CH_2Me), 4.79 and 5.05 (2 H, ABq, *J* 12.2, OCH_2Ph), 6.14 (2 H, d, *J* 7.8, ArH), 6.38 (1 H, s, 2-H), 6.69 (2 H, d, *J* 7.3, ArH), 6.97 (2 H, m, ArH), 7.08 (2 H, m, ArH), 7.22 (5 H, m, ArH) and 7.36 (2 H, m, ArH); δ_C 13.98, 41.74, 47.81, 48.81, 61.60, 64.28, 90.39, 127.50, 127.65, 127.81, 127.94, 128.11, 128.93, 128.98, 129.16, 130.73, 134.63, 135.19, 135.52, 152.19, 166.17, 172.74 and 200.45.

(2'S,4'R)-4'-*Benzyl*-3'-*benzyloxycarbonyl*-5'-*oxo*-2'-*phenyloxazolidin*-4'-*ylacetyl Chloride* **12**.—The acid **10** (402 mg, 0.90 mmol) was dissolved in CH_2Cl_2 (32 cm^3) and the solution was cooled to 0 °C. Freshly distilled oxalyl dichloride (0.39 cm^3, 4.51 mmol, 5 mol equiv.) and a catalytic quantity of DMF were added. The mixture was stirred at 0 °C for 2 h and at 20 °C for 16 h. The solvent was evaporated off, more CH_2Cl_2 (2 cm^3) was added and evaporated off (repeated 3 times). Final traces of oxalyl dichloride were removed at 1 mmHg to yield acid chloride **12** as a beige solid (418 mg, 100%), which was used in subsequent steps without further purification; δ_H 3.20 and 3.51 (2 H, ABq, *J* 13.2, 4-CH_2Ph), 3.61 and 4.37 (2 H, ABq, *J* 19.1, CH_2COCl), 4.84 and 5.10 (2 H, ABq, *J* 12.2, OCH_2Ph), 6.12 (2 H, d, *J* 7.3, ArH), 6.30 (1 H, s, 2-H), 6.72 (2 H, d, *J* 7.8, ArH), 7.01 (4 H, m, ArH), 7.23 (5 H, m, ArH) and 7.37 (2 H, m, ArH).

(2'S,4'R)-(−)-4-(4'-*Benzyl*-3'-*benzyloxycarbonyl*-5'-*oxo*-2'-*phenyloxazolidin*-4'-*yl*)-3-*oxo*-2-(*triphenylphosphoranylidene*)-*butanoate* **13**.—*Method A*. The acid chloride **12** (412 mg, 0.89 mmol, 1 mol equiv.) was dissolved in CH_2Cl_2 (32 cm^3) and the solution was cooled to 0 °C. $Ph_3P{=}CHCO_2Et$ (619 mg, 1.78 mmol, 2 mol equiv.) was added and the solution was stirred at 0 °C for 1.5 h and at 20 °C for 4.5 h. The solvent was evaporated off and the residue was purified by radial chromatography using a 4 mm silica gel chromatotron plate, and elution with light petroleum–ethyl acetate (55:45) to give the *oxazolidinone* **13** as a solid (691 mg, quant), mp 209–211 °C (from ethyl acetate–light petroleum) (Found: C, 74.1; H, 5.4; N, 1.8. $C_{48}H_{42}NO_7P$ requires C, 74.31; H, 5.46; N, 1.81%); $[\alpha]_D^{20}$ −4 (*c* 1.5, CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 1790, 1710, 1666 and 1559; δ_H([$^2H_6$]DMSO; 85 °C) 0.77 (3 H, t, *J* 7.3, Me), 3.26 and 3.53 (2 H, ABq, *J* 13.2, 4-CH_2Ph), 3.37 and 4.74 (2 H, ABq, *J* 17.6, 4-CH_2CO), 3.77 (2 H, m, OCH_2Me), 5.18 (2 H, m, OCH_2Ph), 5.43 (1 H, s, 2-H), 6.16 (2 H, d, *J* 7.3, ArH), 7.10 (4 H, m, ArH), 7.37 (6 H, m, ArH) and 7.68 (13 H, m, ArH); δ_P 18.1; δ_C 13.75; 41.93, 42.74, 45.95 (d, *J* 7.6), 48.01 (d, *J* 7.1), 58.28, 58.54, 65.22, 65.72, 66.63, 67.24, 71.08 (d, *J* 109.3), 71.24 (d, *J* 110.8), 89.35, 89.50, 125.56 (d, *J* 93.7), 125.89 (d, *J* 93.1), 126.88, 127.01, 127.33, 127.51, 127.62, 127.79, 127.91, 127.97, 128.25, 128.45 (d, *J* 12.0), 128.60, 128.62 (d, *J* 12.6), 128.64, 128.70, 129.08, 130.80, 131.63 (d, *J* 2.5), 133.15 (d, *J* 10.1), 133.20 (d, *J* 9.6), 135.40, 135.65, 137.71, 135.98, 136.02, 136.46, 151.69, 152.23, 167.33 (d, *J* 14.1), 167.45 (d, *J* 14.1), 173.96, 174.13, 192.14 (d, *J* 6.0) and 192.22 (d, *J* 5.1).

Method B. The oxazolidinone **8** (100 mg, 0.26 mmol) was dissolved in THF (10 cm^3) and the solution was cooled to −78 °C. LiHMDS (0.28 cm^3, 0.28 mmol of a 1 mol dm^{-3} solution in THF, 1.1 mol equiv.) was added and the resulting yellow solution was stirred at −78 °C for 7 min. $BrCH_2COC(Ph_3)$-CO_2Et[18] (127 mg, 0.27 mmol, 1.05 mol equiv.) was added and the solution was stirred at −78 °C for 2 h and was then allowed to warm to 20 °C over a period of 16 h. The THF was evaporated off and the residue was partitioned between saturated aq. NH_4Cl (10 cm^3) and CH_2Cl_2 (10 cm^3). The aqueous layer was separated, and extracted with CH_2Cl_2 (2 × 10 cm^3). The combined CH_2Cl_2 extracts were dried ($MgSO_4$) and evaporated. Further purification on a 2 mm silica gel chromatotron plate and elution with light petroleum–ethyl acetate (55:45 yielded the oxazolidinone **13** as a solid (52 mg, 26%); δ_H as given above.

(5R)-*Ethyl Hydrogen* 5-*Benzyl*-5-*benzyloxycarbonylamino*-3-*oxo*-2-(*triphenylphosphoranylidene*)*hexanedioate* **17**.—Methanol (48 cm^3) followed by aq. LiOH (24 cm^3 of a 3.33 mol dm^{-3} solution, 79.9 mmol, 103 mol equiv.) were added to a solution of the oxazolidinone **13** (600 mg, 0.77 mmol, 1 mol equiv.) in THF (48 cm^3). The mixture was refluxed for 4 h, cooled to 0 °C and acidified to pH 1–3 (universal indicator paper) with 2 mol dm^{-3} aq. HCl. The THF was evaporated off and the remaining solution was extracted with ethyl acetate (3 × 50 cm^3). The combined extracts were dried ($MgSO_4$) and the solvent was evaporated off at 20 mmHg, and finally at 1 mmHg for 16 h, to give compound **17** as a solid (530 mg, quant), which was used in subsequent steps without further purification [Found: MH^+ (FAB), 688.2461. $C_{41}H_{39}NO_7P$ requires M, 688.2464]; ν_{max}(KBr)/cm^{-1} 3404, 1790, 1715, 1667 and 1559; δ_H 0.74 (3 H, t, *J* 7.1, Me), 2.86 and 3.52 (2 H, ABq, *J* 13.5, 5-CH_2Ph), 2.94 and 5.03 (2 H, ABq, *J* 17.6, CCH_2CO), 3.83 (2 H, m, CH_2Me), 4.97 and 5.29 (2 H, ABq, *J* 12.2, OCH_2Ph), 6.08 (1 H, s, NH), 6.87 (2 H, d, *J* 7.8, ArH), 7.09 (3 H, m, ArH), 7.33–7.51 (11 H, m, ArH), 7.57 (3 H, m, ArH) and 7.69 (6 H, m, ArH); δ_P 18.6; δ_C 13.45, 37.55, 41.49 (d, *J* 6.1), 59.85, 62.64, 65.94, 124.44 (d, *J* 93.7), 126.62, 127.94, 128.05, 128.31, 128.59, 128.82 (d, *J* 13.1), 129.70, 132.40 (d, *J* 2.1), 133.06 (d, *J* 10.0), 135.52, 136.86, 154.26, 166.55 (d, *J* 13.1), 173.94 and 192.79 (d, *J* 4.0).

(2R)-(+)-6-*Ethyl* 1-*Methyl* 2-*Benzyl*-2-*benzyloxycarbonylamino*-4-*oxo*-5-(*triphenylphosphoranylidene*)*hexanedioate* **18**.—The keto acid phosphorane **17** (89 mg, 0.13 mmol) was dissolved in THF (1 cm^3) and the solution was treated with a large excess of freshly distilled CH_2N_2 in diethyl ether. The excess of CH_2N_2 was allowed to evaporate off at 20 °C over a period of 16 h and the residue was purified by radial chromatography using a 1 mm silica gel chromatotron plate, and elution with a gradient of ethyl acetate (25–50%) in light petroleum, to give *compound* **18** (75 mg, 82%) as a solid, mp 181–185 °C (from ethyl acetate–light petroleum (Found: C, 71.6; H, 5.4; N, 1.9. $C_{42}H_{40}NO_7P$ requires C, 71.89; H, 5.75; N, 2.00%); $[\alpha]_D^{20}$ +4 (*c* 3.1, CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 3419, 1722, 1666 and 1555; δ_H 0.69 (3 H, t, *J* 7.1, CH_2*Me*), 3.29 and 3.56 (2 H, ABq, *J* 13.6, CCH_2Ph), 3.49 (3 H, s, OMe), 3.74 (4 H, m, CH_2Me and CCH_2CO), 5.11 and 5.21 (2 H, ABq, *J* 12.7, OCH_2Ph), 6.22 (1 H, s, NH), 6.97 (2 H, m, ArH), 7.14 (3 H, m, ArH), 7.37 (11 H, m, ArH), 7.47 (3 H, m, ArH) and 7.61 (6 H, m, ArH); δ_P 18.0; δ_C 13.73, 40.82, 45.56 (d, *J* 6.1), 52.04, 58.56, 62.13, 65.79, 71.79 (d, *J* 110.8), 126.27 (d, *J* 93.7), 126.50, 127.85, 128.86 (d, *J* 14.1), 128.33, 128.50, 130.33, 131.59 (d, *J* 3.0), 132.22 (d, *J* 10.0), 136.21, 137.15, 154.75, 167.57 (d, *J* 15.1), 173.15 and 193.11 (d, *J* 4.0).

(4',R,Z)-(−)- *and* (+)-(4'R,E)-(+)-*Ethyl* (4'-*Benzyl*-4'-*benzyloxycarbonylamino*-5-*oxotetrahydrofuran*-2'-*ylidene*)*bromoacetate* **19** *and* **20**.—Triethylamine (58 mm^3, 0.44 mmol, 1 mol equiv.) followed by Br_2 (22 mm^3, 0.44 mmol, 1 mol equiv.) were added to a solution of the keto acid phosphorane **17** (300

mg, 0.44 mmol) in CH_2Cl_2 (30 cm^3) at 0 °C. The solution was stirred at 0 °C for 20 min and then at 20 °C for 30 min. The solvent was evaporated off to give the crude (*Z*)- and (*E*)-bromo enolactones (**19** and **20**, respectively) in the ratio 54% *Z*:46% *E*, by ^1H NMR spectroscopy. Purification by radial chromatography using a 2 mm silica gel chromatotron plate, and elution with CH_2Cl_2, gave the (*Z*)-*enolactone* **19** (78 mg, 37%) as a solid, which was used in subsequent steps without further purification (Found: C, 56.75; H, 4.7; N, 2.5. $C_{23}H_{22}BrNO_6$ requires C, 56.57; H, 4.54; N, 2.87%); $[\alpha]_D^{20}$ −2 (*c* 1.5, CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 3335, 1825, 1704, 1638 and 1524; δ_H 1.32 (3 H, t, *J* 7.1, Me), 2.98 and 3.14 (2 H, ABq, *J* 13.2, 4′-CH_2Ph), 3.49 and 3.80 (2 H, ABq, *J* 19.1, 3′-H_2), 4.22 (2 H, m, CH_2Me), 5.09 (2 H, m, OCH_2Ph), 5.40 (1 H, s, NH), 7.17 (2 H, m, ArH) and 7.34 (8 H, m, ArH); δ_C 14.12, 39.07, 42.50, 60.38, 62.07, 67.68, 90.68, 128.33, 128.42, 128.50, 128.61, 129.04, 129.92, 131.63, 135.35, 154.95, 159.71, 162.66 and 172.69. Further elution with CH_2Cl_2 gave the (E)-*enolactone* **20** (66 mg, 31%) as a solid, which was used in subsequent steps without further purification (Found: C, 56.8; H, 4.6; N, 2.8%); $[\alpha]_D^{20}$ +7 (*c* 0.9, CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 3337, 1823, 1712, 1642 and 1523; δ_H 1.29 (3 H, t, *J* 7.1, Me), 3.03 and 3.15 (2 H, ABq, *J* 13.2, 4′-CH_2Ph), 3.37 (2 H, m, 3′-H_2), 4.22 (2 H, q, *J* 7.1, CH_2Me), 5.11 (2 H, m, OCH_2Ph), 5.40 (1 H, s, NH), 7.18 (2 H, m, ArH) and 7.34 (8 H, m, ArH); δ_C 14.09, 40.22, 42.49, 59.94, 62.20, 67.68, 94.60, 128.37, 128.53, 128.62, 129.08, 129.88, 131.71, 135.35, 154.93, 155.25, 160.72 and 173.86.

(4′R,Z)- *and* (4′R,E)-*Ethyl* (4′-*Benzyl*-4′-*benzyloxycarbonylamino*-1′-*ethoxycarbonylmethyl*-5′-*oxopyrrolidin*-2′-*ylidene*)-*bromoacetate* **22** *and* **23**.—Glycine ethyl ester hydrochloride (60 mg, 0.43 mmol, 3 mol equiv.) and triethylamine (57 mm^3, 0.43 mmol, 3 mol equiv.) were added to a solution of the (*E*)-bromo enolactone **20** (70 mg, 0.14 mmol) in CH_2Cl_2 (35 cm^3). The mixture was stirred for 16 h, washed with water (35 cm^3), dried ($MgSO_4$), and the solvent was evaporated off to give the bromo hydroxy lactam **21** as a complex mixture of isomers (250 mg, quant), which was used in subsequent steps without further purification.

Compounds **21** (0.14 mmol) and PTSA (14 mg) were dissolved in 1,2-dichloroethane (35 cm^3) and the solution was refluxed, with azeotropic removal of water, for 3.5 h. The solvent was evaporated off and the residue was purified by radial chromatography using a 1 mm silica gel chromatotron plate, and elution with CH_2Cl_2–ethyl acetate (94:4) to give an inseparable mixture of the (Z)- and (E)-*bromo enamino esters* **22** and **23**, respectively, as a pale yellow oil (~85:15; 52 mg, 65%) [Found: MH^+ (CI), 573.1238. $C_{27}H_{30}{}^{79}BrN_2O_7$ requires MH, 573.1237]; ν_{min}(film)/cm^{-1} 3351, 1747, 1713 and 1602; (*Z*)-isomer **22** from the mixture had δ_H 1.29 (3 H, t, *J* 7.1, Me), 1.33 (3 H, t, *J* 7.1, Me), 3.04 and 3.09 (2 H, ABq, *J* 13.2, 4′-CH_2Ph), 3.42 and 3.92 (2 H, ABq, *J* 17.3, 3′-H_2), 4.25 (4 H, m, 2 × CH_2Me), 4.80 (2 H, br m, NCH_2), 5.08 (3 H, m, OCH_2Ph and =CH), 5.29 (1 H, s, NH), 7.13 (2 H, m, ArH) and 7.34 (8 H, m, ArH); δ_C 14.08, 14.18, 40.31, 42.49, 44.89, 59.09, 61.79, 61.91, 67.16, 98.67, 127.85, 128.34, 128.43, 128.54, 128.70, 130.10, 133.25, 135.74, 148.00, 154.75, 163.55, 167.69 and 176.52; (*E*)-isomer **23** from the mixture had δ_H (selected data) 4.55 (2 H, br m, NCH_2) and 5.36 (1 H, s, NH). Further elution with light petroleum–ethyl acetate (7:3) gave the *imide* **28** as a pale yellow oil which was not purified further (8 mg, 13%) (Found: M^+, 424.1629. $C_{23}H_{24}N_2O_6$ requires M, 424.1634); ν_{max}(film)/cm^{-1} 3350, 1790, 1715, 1630 and 1520; δ_H 1.28 (3 H, t, *J* 7.1, Me), 3.04 (4 H, m, 4′-CH_2Ph and 3′-H_2), 4.22 (4 H, m, CH_2Me and NCH_2), 5.04 and 5.11 (2 H, ABq, *J* 12.2, OCH_2Ph), 5.34 (1 H, s, NH), 7.17 (2 H, m, ArH) and 7.34 (8 H, m, ArH); δ_C 14.06, 39.51, 39.82, 42.62, 60.17, 61.96, 67.42, 128.13, 128.38, 128.49, 128.63, 129.05, 130.06, 132.96, 135.56, 154.97, 166.56, 172.90 and 176.79.

The same sequence using the (*Z*)-bromo enolactone **19**, rather than its *E*-isomer **20**, gave the enamino esters **22** and **23** and the imide **28** in the same yield and isomer ratio.

(4′R,E)-(−)-*Ethyl* (4′-*Benzyl*-4′-*benzyloxycarbonylamino*-5′-*oxotetrahydrofuran*-2′-*ylidene*)*acetate* **24**.—The keto acid phosphorane **17** (62 mg, 0.090 mmol) was dissolved in THF (7 cm^3) and the solution was refluxed for 6 h. The solvent was evaporated off and the residue was purified by radial chromatography using a 1 mm silica gel chromatotron plate, and elution with CH_2Cl_2–ethyl acetate (97:3), to give the *enolactone* **24**, as a pale yellow oil (37 mg, 73%), which crystallised on storage at 4 °C, mp 106–108 °C (from ethyl acetate–light petroleum) (Found: C, 67.7; H, 5.4; N, 3.5. $C_{23}H_{23}NO_6$ requires C, 67.47; H, 5.66; N, 3.42%); $[\alpha]_D^{20}$ −13 (*c* 0.6, CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 3391, 1807, 1712 and 1526; δ_H 1.27 (3 H, t, *J* 7.1, Me), 2.99 and 3.14 (2 H, ABq, *J* 13.2, 4′-CH_2Ph), 3.50 and 3.82 (2 H, ABq, *J* 19.1, 3′-H_2), 4.15 (2 H, m, CH_2Me), 5.10 (2 H, m, OCH_2Ph), 5.34 (1 H, s, =CH), 5.46 (1 H, s, NH), 7.18 (2 H, m, ArH) and 7.32 (8 H, m, ArH); δ_C 14.26, 37.30, 42.53, 59.52, 60.10, 67.68, 97.87, 128.33, 128.41, 128.53, 128.63, 129.08, 130.08, 131.95, 135.42, 154.93, 163.62, 166.25 and 173.72.

(4′R,E)-(+)-*Ethyl* (4′-*Benzyl*-4′-*benzyloxycarbonylamino*-1′-*ethoxycarbonylmethyl*-5′-*oxopyrolidin*-2′-*ylidene*)*acetate* **26**.—*Method A*. Glycine ethyl ester hydrochloride (75 mg, 0.54 mmol, 2 mol equiv.) and triethylamine (71 mm^3, 0.54 mmol, 2 mol equiv.) were added to a solution of the enolactone **24** (110 mg, 0.27 mmol) in CH_2Cl_2 (40 cm^3). The mixture was stirred for 16 h, washed with water (40 cm^3), dried ($MgSO_4$), and evaporated to give the hydroxy lactams **25** as a yellow oil (107 mg, 78%), which was used in subsequent steps without further purification; ν_{max}(film)/cm^{-1} 3412 and 1713; δ_C (selected resonances for both diastereoisomers) 13.95, 13.98, 14.02, 40.70, 41.37, 42.52, 42.80, 42.85, 43.59, 43.64, 59.64, 60.16, 60.65, 61.19, 61.79, 66.69, 67.36, 76.02, 86.38, 126.46, 127.38, 127.47, 127.53, 128.11, 128.19, 128.35, 128.43, 128.50, 128.65, 128.70, 130.44, 134.87, 136.03, 154.77, 155.78, 168.45, 169.06, 169.62, 170.11, 173.68 and 174.21; *m*/*z* (CI) 513 (MH^+, 5%), 495 (13), 403 (22), 108 (14) and 91 (100).

A solution of the hydroxy lactams **25** (100 mg, 0.20 mmol) and PTSA (4 mg) in 1,2-dichloroethane (35 cm^3) was refluxed, with azeotropic removal of water, for 3 h. After cooling to 20 °C the solution was washed with water (10 cm^3), dried ($MgSO_4$), and evaporated. Purification by radial chromatography using a 1 mm silica gel chromatotron plate, and elution with CH_2Cl_2–ethyl acetate (94:4), gave the *enamino ester* **26** as an oil (65 mg, 68%) (Found: C, 65.9; H, 6.4; N, 5.4. $C_{27}H_{30}N_2O_7$ requires C, 65.57; H, 6.11; N, 5.66%); $[\alpha]_D^{20}$ +20 (*c* 2.3, CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 3345, 1745, 1709, 1630 and 1520; δ_H 1.28 (3 H, t, *J* 7.1, Me), 1.28 (3 H, t, *J* 7.1, Me), 3.05 (2 H, m, 4′-CH_2Ph), 3.38 and 3.89 (2 H, ABq, *J* 18.6, 3′-H_2), 4.09 and 4.43 (2 H, ABq, *J* 17.6, NCH_2), 4.15 (2 H, m, CH_2Me), 4.22 (2 H, q, *J* 7.1, CH_2Me), 4.99 (1 H, s, =CH), 5.01 and 5.10 (2 H, ABq, *J* 11.8, OCH_2Ph), 5.27 (1 H, s, NH), 7.17 (2 H, m, ArH) and 7.31 (8 H, m, ArH); δ_C 14.04, 14.34, 36.90, 41.94, 42.46, 59.33, 59.73, 61.93, 67.14, 92.86, 127.75, 128.30, 128.39, 128.51, 128.75, 130.12, 133.32, 135.74, 154.65, 154.81, 166.38, 166.52 and 175.56.

Method B. $TiCl_4$ (4 mm^3, 0.037 mmol, 0.5 mol equiv.) was added to compound **11** (35 mg, 0.068 mmol) and glycine ethyl ester[19] (68 mg, 0.66 mmol, 10 mol equiv.) in a mixture of diethyl ether (1 cm^3) and toluene (1 cm^3), at 0 °C. The solution, which turned orange-brown upon addition of $TiCl_4$, was allowed to warm to 20 °C and was then refluxed for 18 h. The solvent was

evaporated off and the residue was purified by preparative TLC on silica and elution with CH_2Cl_2–ethyl acetate (98:2) to give the enamino ester **26** as a pale yellow oil (4 mg, 12%); δ_H as given earlier.

(4'R,E)-(+)-*Ethyl* (4'-*Benzyl*-4'-*benzyloxycarbonylamino*-1'-*ethoxycarbonylmethylcarbamoylmethyl*-5'-*oxopyrrolidin*-2'-*ylidene*)*acetate* **34**.—*Method A*. Glycylglycine ethyl ester hydrochloride (78 mg, 0.40 mmol, 5.4 mol equiv.) and triethylamine (52 mm^3, 0.40 mol equiv.) were added to a solution of the enolactone **24** (30 mg, 0.073 mmol, 1 mol equiv.) in 1,2-dichloroethane (10 cm^3) and the mixture was refluxed, with azeotropic removal of water, for 44 h. After cooling to 20 °C, the mixture was washed with water (10 cm^3), dried ($MgSO_4$), and evaporated to give a yellow oil (43 mg), which was dissolved in 1,2-dichloroethane (10 cm^3). PTSA (16 mg) was added and the solution was refluxed, with azeotropic removal of water, for 4 h. The solvent was evaporated off and the residue was purified by radial chromatography using a 1 mm silica gel chromatotron plate, and elution with CH_2Cl_2–ethyl acetate (4:1), to give the *enamino ester* **34** as an oil (26 mg, 64%); $[\alpha]_D^{20}$ +4 (*c* 0.8, CH_2Cl_2) (Found: M^+, 551.2258. $C_{29}H_{33}N_3O_8$ requires M, 551.2268); ν_{max}(film)/cm^{-1} 3339, 1748, 1694, 1633 and 1538; δ_H 1.21 (3 H, t, *J* 7.1, Me), 1.25 (3 H, t, *J* 7.1, Me), 2.97 and 3.11 (2 H, ABq, *J* 13.2, 4'-CH_2Ph), 3.37 (1 H, dd, *J* 2.0 and 19.1, 3'-H^a), 3.65 and 4.67 (2 H, ABq, *J* 17.1, NCH_2), 3.79 (1 H, dd, *J* 1.5 and 19.1, C'-H^b), 3.84 (1 H, dd, *J* 5.9 and 17.3, NCH^a), 4.02 (1 H, dd, *J* 5.9 and 17.3, NHC*H*^{b}), 4.12 (2 H, q, *J* 7.1, CH_2Me), 4.14 (2 H, q, *J* 7.1, CH_2Me), 5.02 (2 H, s, OCH_2Ph), 5.11 (1 H, s, =CH), 5.42 (1 H, s, CBzN*H*), 7.19 (2 H, m, ArH), 7.29 (8 H, m, ArH) and 7.43 (1 H, br t, N*H*CH_2); δ_C 14.05, 14.26, 36.87, 41.37, 42.22, 44.08, 59.10, 59.77, 61.15, 67.69, 93.42, 128.10, 128.19, 128.53, 128.65, 128.97, 130.00, 131.51, 135.31, 153.87, 155.45, 166.10, 166.68, 168.88 and 175.53.

Method B. Glycine *tert*-butyl ester hydrochloride (13 mg, 0.078 mmol, 2 mol equiv.) and triethylamine (10 mm^3, 0.078 mmol, 2 mol equiv.) were added to a solution of the enolactone **24** (16 mg, 0.039 mmol, 1 mol equiv.) in CH_2Cl_2 (15 cm^3). The mixture was stirred for 16 h at 20 °C, washed with water (15 cm^3), dried ($MgSO_4$), and evaporated to yield the hydroxy lactams **35** as an oil (21 mg, 100%), which was used in subsequent steps without further purification [Found: (M − 18)$^+$, 522.2375. $C_{29}H_{34}N_2O_8$ requires *m*/*z* 522.2368]; ν_{max}(film)/cm^{-1} 3412 and 1711; δ_H 1.25 (3 H, t, *J* 7.1, CH_2Me), 1.49 (9 H, s, Bu^t), 2.78 (4 H, m, 3'-H_2 and 2'-CH_2CO_2Et), 3.15 and 3.32 (2 H, ABq, *J* 13.7, 4'-CH_2Ph), 3.92 and 4.22 (2 H, ABq, *J* 10.8, NCH_2), 4.17 (2 H, m, CH_2Me), 5.02 and 5.09 (2 H, ABq, *J* 12.2, OCH_2Ph), 5.30 (1 H, s, NH), 7.20 (2 H, m, ArH) and 7.34 (8 H, m, ArH); δ_C 14.04, 27.96, 42.40, 42.61, 43.81, 60.21, 61.21, 66.71, 82.95, 86.45, 127.47, 128.18, 128.50, 128.66, 130.50, 135.03, 136.14, 154.82, 169.14, 170.06 and 174.20.

Compounds **35** (21 mg, 0.039 mmol) and PTSA (2 mg) were dissolved in 1,2-dichloroethane (10 cm^3) and the solution was refluxed, with azeotropic removal of water, for 3 h. Evaporation of the solvent gave compound **36** as a beige oil (22 mg), which was used subsequently without further purification; δ_H 1.27 (3 H, t, *J* 7.1, CH_2*Me*), 1.47 (9 H, s, Bu^t), 3.05 (2 H, m, 4'-CH_2Ph), 3.37 (1 H, d, *J* 17.6, 3'-H^a), 3.82–4.31 (5 H, m, CH_2Me, 3'-H^b and NCH_2), 5.08 (2 H, m, OCH_2Ph), 5.29 (1 H, s, =CH), 5.38 (1 H, s, NH), 7.17 (2 H, m ArH) and 7.31 (8 H, m, ArH).

The *tert*-butyl enamino ester **36** (0.039 mmol), PTSA (2 mg) and benzene (10 cm^3) were refluxed together, with azeotropic removal of water, for 3 h. Evaporation of the solvent yielded a brown oil (23 mg), used subsequently without further purification, containing *enamino ester* **37** (Found: M^+, 466.1737. $C_{25}H_{26}N_2O_7$ requires M, 466.1740); δ_H 1.27 (3 H, t, *J* 7.1, Me), 3.01 and 3.07 (2 H, ABq, *J* 13.2, 4'-CH_2Ph), 3.37 and 3.84 (2 H, ABq, *J* 18.6, 3'-H_2), 4.15 (2 H, m, CH_2Me), 4.23 and 4.36 (2 H, ABq, *J* 17.5, NCH_2), 5.03 and 5.07 (2 H, ABq, *J* 12.2, OCH_2Ph), 5.06 (1 H, s, =CH), 5.38 (1 H, s, NH), 7.17 (2 H, m, ArH) and 7.37 (8 H, m, ArH).

The acid **37** (0.035 mmol), DCC (7 mg, 0.035 mmol, 1 mol equiv.), glycine ethyl ester hydrochloride (5 mg, 0.040 mmol, 1.1 mol equiv.) and triethylamine (5 mm^3, 0.040 mmol, 1.1 mol equiv.) were stirred in CH_2Cl_2 (2 cm^3) for 16 h at 20 °C. The mixture was diluted with CH_2Cl_2 (5 cm^3), washed with water (7 cm^3), dried ($MgSO_4$), and evaporated. Purification by radial chromatography using a 1 mm silica gel chromatotron plate, and elution with ethyl acetate–CH_2Cl_2 (4:1), gave the enamino ester **34** (15 mg). Identical data with those given above.

(3'R,2S,E)-(−)-*Methyl* 2-(3'-*Benzyl*-3'-*benzyloxycarbonylamino*-5'-*ethoxycarbonylmethylene*-2'-*oxopyrrolidin*-1'-*yl*)*propanoate* **39**.—(*S*)-Alanine methyl ester hydrochloride (189 mg, 1.36 mmol, 15 mol equiv.) and triethylamine (179 mm^3, 1.36 mmol, 15 mol equiv.) were added to a solution of the enolactone **24** (37 mg, 0.090 mmol) in 1,2-dichloroethane (25 cm^3) and the mixture was refluxed, with azeotropic removal of water, for 43 h. The solvent was evaporated off and the residue was purified by radial chromatography using a 1 mm silica gel chromatotron plate, and elution with CH_2Cl_2–ethyl acetate (95:5), to give *compound* **39** as a yellow oil (35 mg, 78%) [Found: (M + K), 533.1692. $C_{27}H_{30}N_2O_7K$ requires *m*/*z*, 533.1690]; $[\alpha]_D^{20}$ −17 (*c* 1.0, CH_2Cl_2); ν_{max}(film)/cm^{-1} 3341, 1743, 1712, 1625 and 1522; δ_H 1.27 (3 H, t, *J* 7.1, CH_2*Me*), 1.47 (3 H, d, *J* 7.3, NCHMe), 2.98 and 3.07 (2 H, ABq, *J* 13.2, 3'-CH_2Ph), 3.37 (1 H, d, *J* 18.6, 4'-H^a), 3.67 (3 H, s, OMe), 3.71 (1 H, dd, *J* 2 and 18.6, 4'-H^b), 3.87 (1 H, q, *J* 7.3, NCH), 4.15 (2 H, m, CH_2Me), 5.00 (4 H, m, OCH_2Ph, NCH and =CH), 5.27 (1 H, br s, NH), 7.17 (2 H, m, ArH) and 7.33 (8 H, m, ArH); δ_C 12.71, 14.34, 36.99, 42.37, 49.45, 52.72, 59.05, 59.73, 67.12, 93.70, 127.69, 128.22, 128.31, 128.55, 128.78, 130.30, 133.09, 135.75, 153.42, 154.73, 166.64, 169.76 and 175.36. The ^{13}C NMR spectrum indicated the presence of < 5% of another diastereoisomer.

(α'R,2R,4S)-(+)-*Benzyl*-4-*Benzyl*-4-{N-[1-(1-*naphthyl*)-*ethyl*]*carbamoylmethyl*}-5-*oxo*-2-*phenyloxazolidine*-3-*carboxylate* **44**.—(*R*)-(+)-1-(1-Naphthyl)ethylamine (52 mm^3, 0.322 mmol, 1 mol equiv.) and *N*-hydroxybenzotriazole·H_2O (50 mg, 0.326 mmol, 1 mol equiv.) were added to a solution of the carboxylic acid **43** (144 mg, 0.323 mmol, prepared from (*R*)-phenylalanine **40** *via* lactone **41** and diester **42** as described for compound **10**) in CH_2Cl_2 (0.65 cm^3) at 0 °C under N_2. The solution was stirred at 0 °C for 10 min after which time DCC (67 mg, 0.325 mmol, 1 mol equiv.) was added and the mixture was stirred for a further 15 min at 0 °C, then at room temp. for 17 h. The reaction mixture was filtered and the filtrate was washed successively with 5% aq. HCl (20 cm^3) followed by water (2 × 20 cm^3). The organic phase was dried (Na_2SO_4) and the residue was filtered off, and chromatographed using a 1 mm silica gel chromatotron plate, and elution with ethyl acetate–light petroleum (33:67) to give *compound* **44** (136 mg, 70%) as an oil (Found: C, 75.1; H, 6.2; N, 5.0. $C_{38}H_{34}N_2O_5 \cdot 1/4H_2O$ requires C, 75.10; H, 5.80; N, 4.61%); $[\alpha]_D^{20}$ +16 (CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 3346, 1793, 1711, 1662 and 1536; δ_H ([$^2H_6$]DMSO, 80 °C) 1.61 (3 H, d, *J* 6.8, Me), 3.04 and 3.63 (2 H, ABq, *J* 15.7, 4-CH_2CO), 3.29 and 3.51 (2 H, ABq, *J* 13.7, 4-CH_2Ph), 5.17 (2 H, br, OCH_2Ph), 5.84 (1 H, quin, *J* 6.8, NC*H*Me), 6.33 (1 H, s, 2-H), 6.42 (2 H, d, *J* 7.3, ArH), 7.09–7.68 (16 H, m, ArH), 7.93 (1 H, d, *J* 7.8, ArH), 8.03 (1 H, d, *J* 7.3, ArH), 8.21 (1 H, d, *J* 8.3, ArH) and 8.70 (1 H, d, *J* 7.3, ArH).

(α'R,2S,4R)-(+)-*Benzyl* 4-*Benzyl*-4-{N-[1-(1-*naphthyl*)*ethyl*]*carbamoylmethyl*}-5-*oxo*-2-*phenyloxazolidin*-3-*carboxylate* **45**.—Compound **45** was prepared from the carboxylic acid **10** as described above for its diastereoisomer **44** (Found: M^+, 598.2474. $C_{38}H_{34}N_2O_5$ requires M, 598.246 75); $[\alpha]_D^{20}$ +64 (CH_2Cl_2); ν_{max}(KBr)/cm^{-1} 3343, 1793, 1711, 1666 and 1536; δ_H([$^2H_6$]DMSO, 85 °C) 1.64 (3 H, d, *J* 6.8, Me), 3.08 and 3.62 (2 H, ABq, *J* 16.1, 4-CH_2CO), 3.29 and 3.49 (2 H, ABq, *J* 13.7, 4-CH_2Ph), 4.64 (1 H, br, OCH_2Ph), 4.99 (1 H, d, *J* 12.7, OCH_2Ph), 5.86 (1 H, quin, *J* 6.8, C*H*Me), 6.11 (1 H, s, 2-H), 6.29 (2 H, d, *J* 6.9, ArH), 6.88 (1 H, br, ArH), 7.04–7.70 (15 H, m, ArH), 7.92 (1 H, d, *J* 7.8, ArH), 8.03 (1 H, d, *J* 7.8, ArH), 8.27 (1 H, d, *J* 8.3, ArH) and 8.69 (1 H, d, *J* 6.9, ArH).

X-Ray Crystallographic Determination for Compound **10**.—Single-crystal data collection was performed at 130 K with Siemens P4 four-circle diffractometer using graphite-monochromatised Mo-Kα radiation (λ = 0.710 73 Å). A thin needle-shaped crystal with dimensions 0.80 × 0.22 × 0.08 mm was used. The compound $C_{26}H_{23}NO_6$, M_r = 445.45, crystallised from ethyl acetate–light petroleum in the orthorhombic system, space group $P2_12_12_1$, a = 7.348(1), b = 17.588(4), c = 17.641(4) Å, $\alpha = \beta = \gamma = 90°$, V = 2279.9(8) Å^3, Z = 4, D_{calc} = 1.298 g cm^{-3}, μ(Mo-Kα) = 0.093 mm^{-1}. The unit-cell parameters were determined by least-squares refinements of 19 accurately centred reflections ($10 < 2\theta < 17.5°$). 1312 Unique reflections were collected by adaptive ω scan mode (peak top ω scans of 0.8° with 0.8° offset to background from peak position), ω scan speed 29.6 deg min^{-1}. Of those, 874 were considered as observed according to the criterion $|F| > 4\sigma(F)$. The structure was solved by direct methods by using the SHELXS-86 program.[20] Full-matrix least-squares refinement on F^2 and all subsequent calculations were performed using SHELXL-93 program system.[21] The refinement converged with R = 0.0538 and R_w = 0.1029. Tables of non-hydrogen-atom coordinates, bond lengths, bond angles, hydrogen-atom coordinates and anisotropic thermal parameters have been deposited with the Cambridge Crystallographic Data Centre (CCDC).‡

Acknowledgements

This work was partially supported by research grant from the Foundation for Research Science and Technology (New Zealand). We thank Professor W. T. Robinson (Department of Chemistry, University of Canterbury, New Zealand) for help with the X-ray crystallography.

‡ See 'Instructions for Authors', in the January issue.

References

1 M. W. Holladay and A. M. Nadzan, *J. Org. Chem.*, 1991, **56**, 3900 and references therein; D. H. Rich, in *Comprehensive Medicinal Chemistry*, ed. C. Hansch, P. G. Sammes and J. B. Taylor, Pergamon Press, New York, 1990; vol. 2, pp. 404–406; K. A. Newlander, J. F. Callahan, M. L. Moore, T. A. Tomaszek, Jr. and W. F. Hoffman, *J. Med. Chem.*, 1993, **36**, 2321; S. E. de Laszlo, B. L. Bush, J. J. Doyle, W. J. Greenlee, D. G. Hangauer, T. A. Halgren, R. J. Lynch, T. W. Schorn and P. K. S. Siegl, *J. Med. Chem.*, 1992, **35**, 833; S. Thaisrivongs, D. T. Pals, S. R. Turner and L. T. Kroll, *J. Med. Chem.*, 1988, **31**, 1369.
2 R. B. Silverman, *Mechanism-Based Enzyme Inactivation: Chemistry and Enzymology*, CRC Press, Florida, 1988, vol. 1; A. D. Abell, in *Detailed Reaction Mechanisms: Mechanisms of Biological Importance*, ed. J. M. Coxon, JAI Press, London, 1992, vol. 2, pp. 243–260.
3 M. Sandler and H. J. Smith, *Design of Enzyme Inhibitors as Drugs*, Oxford University Press, 1989.
4 (*a*) S. B. Daniels and J. A. Katzenellenbogen, *Biochemistry*, 1986, **25**, 1436; (*b*) M. J. Sofia and J. A. Katzenellenbogen, *J. Med. Chem.*, 1968, **29**, 230; (*c*) S. Naruto, I. Motoc, G. R. Marshall, S. B. Daniels, M. J. Sofia and J. A. Katzenellenbogen, *J. Am. Chem. Soc.*, 1985, **107**, 5262.
5 D.-J. Baek, P. E. Reed, S. B. Daniels and J. A. Katzenellenbogen, *Biochemistry*, 1990, **29**, 4305.
6 A. D. Abell and J. M. Taylor, *J. Org. Chem.*, 1993, **58**, 14.
7 D. Seebach and A. Fadel, *Helv. Chim. Acta*, 1985, **68**, 1243.
8 S. Karaday, J. S. Amato and L. M. Weinstock, *Tetrahedron Lett.*, 1984, **25**, 4337.
9 For a review see: R. M. Williams, *Synthesis of Optically Active α-Amino Acids*, ed. J. E. Baldwin and P. D. Magnus, Pergamon Press, Oxford, 1989, pp. 62–87 and references therein.
10 A. Fadel and J. Salaün, *Tetrahedron Lett.*, 1987, **28**, 2243; M. Gander-Coquoz and D. Seebach, *Helv. Chim. Acta*, 1988, **71**, 224.
11 A. D. Abell and M. D. Oldham, unpublished work.
12 A. D. Abell and J. O. Trent, *J. Chem. Soc., Chem. Commun.*, 1989, 409.
13 A. D. Abell, D. A. Hoult, K. M. Morris, J. M. Taylor and J. O. Trent, *J. Org. Chem.*, 1993, **58**, 1531.
14 A. D. Abell, M. D. Oldham and J. M. Taylor, *J. Org. Chem.*, accepted for publication.
15 M. M. Kayser, K. L. Hatt and H. Yu, *Can. J. Chem.*, 1993, **71**, 1010; A. D. Abell, K. B. Morris and J. C. Litten, *J. Org. Chem.*, 1990, **55**, 5217; A. D. Abell, B. M. Clark and W. T. Robinson, *Aust. J. Chem.*, 1988, **41**, 1243; A. D. Abell, I. R. Doyle and R. A. Massy-Westropp, *Aust J. Chem.*, 1982, **35**, 2277.
16 R. J. P. Cannell, S. J. Kellam, A. M. Owsianka and J. M. Walker, *Planta Med.*, 1988, **54**, 10.
17 R. E. Strube, *Org. Synth.*, Coll. Vol. IV, John Wiley and sons, New York, 1963, 417.
18 V. N. Listvan and A. V. Dombrovskii, *Zh. Obshch. Khim.*, 1968, **38**, 601.
19 M. Bodanszky and A. Bodanszky, *The Practice of Peptide Synthesis*, Springer-Verlag, Berlin, 1984, p. 106.
20 G. M. Sheldrick, *Program for Crystal Structure Solution*, University of Göttingen, 1986.
21 G. M. Sheldrick, *Program for Crystal Structure Solution*, University of Göttingen, 1993.

Paper 4/05645K
Received 16*th September* 1994
Accepted 20*th October* 1994