Supplementary Material (ESI) for Chemical Communications

Penicillins as β-Lactamase-Dependent Prodrugs: Enabling Role of a Vinyl Ester Exocyclic to the Lactam Ring

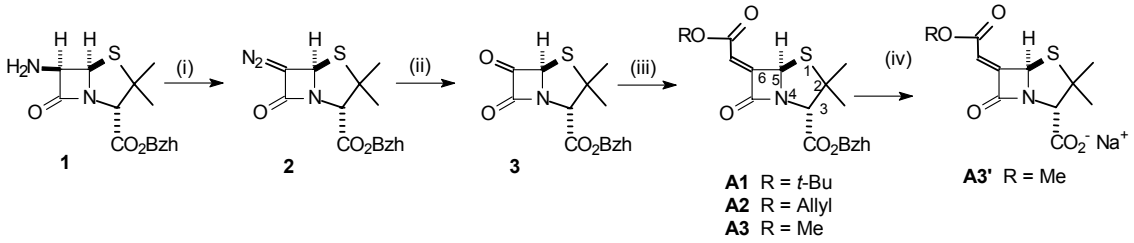
Carol C. Ruddle and Timothy P. Smyth*

Department of Chemical and Environmental Sciences, University of Limerick, National Technological Park, County Limerick, Ireland. E-mail: timothy.smyth@ul.ie

Supporting Information.

Synthesis of the Penicillin Derivatives.

The penicillins **A1** and **A3** were prepared according to the method of Buynak[1] using commercially available Wittig reagents. The derivatives were obtained exclusively as the (*Z*) geometric isomers.[2]



Scheme S1. (i) Isoamyl nitrite, TFA; (ii) Rhodium (II) octanoate dimer, propylene oxide; (iii) $ROC(O)CH=P(Ph)_3$; (iv) $AlCl_3$, $NaHCO_3$, freeze dry.

Experimental.

Benzhydryl-6-aminopenicillanate (1).

The *p*-toluenesulphonic acid salt of benzyhydryl-6-aminopenicillanate was prepared by the method of Petursson and Waley;[3] mp 155-156 °C (lit.[3] 155-156). This salt (1.589 g, 2.86 mmol) was suspended in dichloromethane (39 mL) and triethylamine (1.86 mL, 5.66 mmol) was added dropwise, at which point no more suspended solid was observed. The solution was stirred for 30 min after which time it was washed with water (3 x 40 mL). The organic layer was dried, filtered and concentrated under reduced pressure to yield **1** as a faintly yellow oily material (1.052g, 2.75 mmol, 96%).

Benzhydryl-6-oxopenicillinate (3).[1a, b]

To a solution of benzhydryl 6-aminopenicillanate **(1)** (1.052 g, 2.75 mmol) in dichloromethane (12 mL) was added isoamyl nitrite (555 µl, 4.0 mmol) and trifluoroacetic acid (12 µl, 0.15 mmol). The reaction mixture was allowed to stir for 30 min. Analysis by TLC (50/50 ethyl acetate/hexane) indicated disappearance of starting material and the formation of a new product of higher rf value. The reaction mixture was diluted with dichloromethane (30 mL) and was washed with water (40 mL). The organic layer was separated, dried and concentrated to dryness under reduced pressure to yield a pale yellow glassy solid. This was dissolved in benzene (13 mL) and propylene oxide (21 mL, 0.3 mol) and rhodium octanoate (~5 mg) were added. The resulting solution was stirred under nitrogen for 15 min during which time the evolution of nitrogen was observed and the colour of the solution darkened. Anaylsis by TLC (50/50, ethyl acetate/hexane) indicated the complete disappearance of the diazo-compound with the appearance of the product and one other component. The reaction mixture was concentrated under vacuum to yield a pale yellow glassy solid (1.055 g); the ^{1}H NMR spectrum of this material was consistent with the literature data for **3**[1 b] without any gross impurities.

Benzhydryl-6-(*Z*)-*t*-butoxycarbonylmethylenepenicillanate (A1)

Benzhydryl-6-oxopenicillinate (**3**) (1.04 g, 2.73 mmol) was dissolved in dichloromethane (13 mL). The solution was cooled to –55 ^{o}C under nitrogen. To this cooled solution *t*-butyl (triphenylphosphoranylidene) acetate (967 mg, 2.57 mmol) in dichloromethane (30 mL) was added dropwise over 30 min. Stirring was continued for a further 10 min. Analysis by TLC (50:50, ethyl acetate/hexane) indicated disappearance of the oxo-compound and the formation of a new product of higher rf value. The solution was allowed warm to room temperature and was washed with water (20 mL), the organic layer was separated, dried and concentrated under reduced pressure to leave crude **A1** as a yellow-green oil. Purification by chromatography (silica gel, hexane/ethyl acetate, 70/30) yielded **A1**[1d] as a yellow-green glassy gel (654 mg, 1.36 mmol, 50%)); (Found C, 67.47; H, 6.28; N, 2.92%.

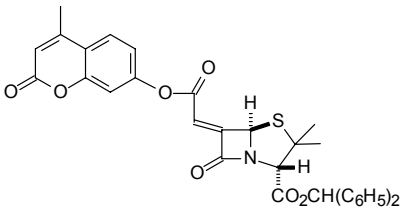
$C_{27}H_{29}NO_5S$ requires C, 67.62; H, 6.09; N, 3.03%); ν_{max} (neat)/cm^{-1} 1781 (lactam), 1744, 1723; δ_H (300 MHz, $CDCl_3$) 1.29 (s, 3H, αC**H**$_3$), 1.51 (s, 9H, C(C**H**$_3$)$_3$), 1.57 (s, 3H, βC**H**$_3$), 4.65 (s, 1H, **H**-3), 5.99 (s, 1H, **H**-5), 6.19 (s, 1H, sidechain C(O)C**H**=), 6.95 (s, 1H, C**H**(Ar)$_2$), 7.20-7.40 (s, 10H, Ar**H**); δ_C (75.47 MHz, $CDCl_3$) δ 25.50 (α**C**H$_3$), 28.07 (C(**C**H$_3$)$_3$), 33.75 (β**C**H$_3$), 63.95, 69.16, 70.71 (**C**-2, **C**-3, **C**-5), 78.45 (**C**H(Ar)$_2$, 82.76 (O**C**(CH$_3$)$_3$), 118.10 (sidechain C(O)**C**H=), 127.07, 127.50, 128.20, 128.38, 128.58, 128.64 (**C**, Ar), 139.15, 139.21 (**C**, Ar), 155.16 (**C**-6),[4] 162.84, 166.69, 166.75 (3 × **C**=O).

Benzhydryl-6-(*Z*)-allyloxycarbonylmethylenepenicillanate (A2)
As for preparation of **A1** but using allyl (triphenylphosphoranylidene)acetate (926 mg, 2.57 mmol). Purification by chromatography (silica gel, hexane/ethyl acetate 60/40) yielded **A2** as a very pale yellow oil (787.9 mg, 1.70 mmol, 66%); ESI-HRMS *m/z* 486.1344 (M + Na^+, $C_{26}H_{25}NO_5S$ requires 486.1346); ν_{max} (neat)/cm^{-1} 1779 (lactam), 1745, 1730; λ_{max} (MeOH)/nm 219, (ε/M^{-1} cm^{-1} 25,455); δ_H (300 MHz, $CDCl_3$) 1.27 (s, 3H, αC**H**$_3$), 1.56 (s, 3H, βC**H**$_3$), 4.66 (s, 1H, **H**-3) overlapping with 4.69 (app. tt, *J* = 5.0, 1.2 Hz, 2H, C**H**$_2$CH=CH$_2$), 5.31 (app. dq, *J* = 16.5, 1.2 Hz, 1H, CH$_2$CH=C**H**H), 5.35 (app. dq, *J* = 23.4, 1.2 Hz, 1H, CH$_2$CH=CH**H**), 5.94 (ddt, *J* = 16.5, 9.6, 1.2 Hz, 1H, CH$_2$C**H**=CH$_2$), 6.04 (d, *J* = 0.5 Hz, 1H, **H**-5), 6.32 (d, *J* = 0.6 Hz, 1H, sidechain C(O)C**H**=), 6.96 (s, 1H, C**H**(Ar)$_2$), 7.27-7.40 (m, 10H, Ar**H**); δ_C (75.47 MHz, $CDCl_3$) 25.37 (α**C**H$_3$), 33.74 (β**C**H$_3$), 63.88, 66.25, 69.11, 70.67 (**C**-2, **C**-3, **C**-5, **C**H$_2$-CH=CH$_2$), 78.45 (**C**H(Ar)$_2$), 115.59, 119.43 (sidechain C(O)**C**H=, CH$_2$-CH=**C**H$_2$), 127.04, 127.53, 128.21, 128.40, 128.59, 128.64 (**C**, Ar), 131.24 (CH$_2$**C**H=CH$_2$), 139.04, 139.11 (**C**, Ar), 156.91 (**C**-6),[4] 163.42, 166.26, 166.67 (3 × **C**=O).

Benzhydryl-6-(*Z*)-methoxycarbonylmethylenepenicillanate (A3)
As for preparation of **A1** but using methyl (triphenylphosphoranylidene)acetate (859 mg, 2.57 mmol). Purification by chromatography (silica gel, hexane/ethyl acetate 70/30) yielded **A3** as a faintly yellow waxy oil (752 mg, 1.72 mmol, 67%); (Found C, 65.98; H, 5.19; N, 3.30%. $C_{24}H_{23}NO_5S$ requires C, 65.89; H, 5.30; N, 3.20%); ν_{max} (neat)/cm^{-1} 1779 (lactam), 1737, 1731; δ_H (300 MHz, $CDCl_3$) 1.26 (s, 3H, αC**H**$_3$), 1.56 (s, 3H, βC**H**$_3$), 3.81 (s, 3H, C**H**$_3$O), 4.66 (s, 1H, **H**-3), 6.03 (s, 1H, **H**-5), 6.31 (s, 1H, sidechain C(O)C**H**=), 6.96 (s, 1H, C**H**(Ar)$_2$), 7.27-7.40 (m, 10H, Ar**H**); δ_C (75.47 MHz, $CDCl_3$) 25.40 (α**C**H$_3$), 33.61 (β**C**H$_3$), 52.46 (**C**H$_3$O), 63.90, 69.03, 70.68 (**C**-2, **C**-3, **C**-5), 78.45 (**C**H(Ar)$_2$), 115.43 (sidechain C(O)**C**H=), 127.07, 127.56, 128.23, 128.40, 128.60, 128.65 (**C**, Ar), 139.05, 139.13 (**C**, Ar), 156.79 (**C**-6),[4] 164.24, 166.30, 166.70 (3 × **C**=O).

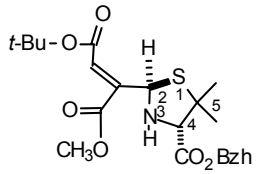
Sodium-6-(*Z*)-methoxycarbonylmethylenepenicillanate (A3')
A3 (263 mg, 0.60 mmol) was dissolved in dichloromethane (13 mL) and was cooled under nitrogen to –84 °C. A solution of aluminium trichloride (198.7 mg, 1.49 mmol) in nitromethane (1.24 mL) and dichloromethane (2.87 mL) was added in one portion to the cooled penicillin solution at which point the solution became intensely yellow. After stirring for 20 min, ethyl acetate (62 mL) and 5% sodium hydrogen carbonate were added successively whilst maintaining the temperature at –84 °C. The resulting slushy mixture was allowed to reach room temperature, the two layers were separated and the aqueous layer was filtered through celite until clear. Ethyl acetate (30 mL) was layered on top of the aqueous filtrate, the pH of this was adjusted to 2.2, and extracted with the ethylacetate. The aqueous layer was extracted with a second portion of ethyl acetate. The organic extracts were combined and extracted with 5% sodium hydrogen carbonate (2 × 50 mL). The combined aqueous extracts were, acidified to pH 2.2 and extracted once more with ethyl acetate (2 × 30 mL). The ethyl acetate was separated, dried and removed under reduced pressure to leave the free acid of **A3'** as a pale yellow glassy solid (122 mg, 75%); ESI-HRMS (MeOH) *m/z* [M - H]$^-$ calcd for $C_{10}H_{11}NO_5S$ 270.0436, found 270.0443; ν_{max} (KBr)/cm^{-1} 3493, 1775 (lactam), 1725 (br); δ_H (300 MHz, $CDCl_3$/DMSO-d6) 1.59 (s, 3H, C**H**$_3$), 1.62 (s, 3H, C**H**$_3$), 3.82 (s, 3H, C**H**$_3$O), 4.59 (s, 1H, **H**-3) 6.02 (s, 1H, **H**-5), 6.33 (s, 1H, sidechain C(O)C**H**=), 7.36 (br s, 1H, CO_2H); δ_C (75.47 MHz, $CDCl_3$) 25.99 (α**C**H$_3$), 32.63 (β**C**H$_3$), 52.54 (**C**H$_3$O), 63.77, 68.70, 70.51 (**C**-2, **C**-3, **C**-5), 115.88 (sidechain C(O)**C**H=), 156.0 (**C**-6), 164.20, 166.46, 172.14 (3 × **C**=O). The free acid (101 mg, 0.372 mmol) was dissolved in ethyl acetate and extracted with aqueous sodium hydrogen carbonate (25.0 mg, 0.297 mmol) and the resulting aqueous layer was lyophilised to leave **A3'** as a yellow solid (80.9 mg, 74%); δ_H (90 MHz, D_2O buffer, pD 7.2) 1.59 (s, 3H, α/βCH$_3$), 1.60 (s, 3H, α/βC**H**$_3$), 3.81 (s, 3H, C**H**$_3$O), 4.37 (s, 1H, **H**-3) 6.04 (s, 1H, **H**-5), 6.45 (s, 1H, sidechain C(O)C**H**=).

Benzhydryl-6-(*Z*)-(methylumbelliferyl)carbonylmethylenepenicillanate (A4)



To a solution of **A2** (227 mg, 0.490 mmol) in THF (5mL) was added a solution of $Pd(PPh_3)_4$ (29 mg, 0.025 mmol) in THF (1.6mL). A solution of toluene-4-sulphinic acid sodium salt (tetrahydrate) (146.0 mg, 0.583 mmol) in water (1.1mL) was added and reaction progress was monitored using TLC (50:50 ethyl acetate/hexane). Three further portions of $Pd(PPh_3)_4$ (2 x 29 and 1 x 10 mg) were added over a period of 50 min in order to obtain complete conversion of the starting material. The reaction mixture was diluted with diethyl ether (20 mL) and water (5 mL). The yellow aqueous layer was retained and the orange ether layer was extracted with water (2 x 5 mL). The aqueous extracts were combined and ethyl acetate (30 ml) was layered on top. The pH was adjusted to 2.2 and the ethyl acetate layer was separated. The aqueous layer was re-extracted with ethyl acetate (2 x 30 mL) and the organic extracts were combined, dried and concentrated to give the free acid of **A2** as a yellow solid (159.5 mg, 0.377 mmol, 77 %); ESI-HRMS (MeOH) *m/z* $[M - H]^-$ calcd for $C_{23}H_{20}NO_5S$ 422.1062, found 422.1061; ν_{max} (KBr)/cm^{-1} 3459, 1779 (lactam), 1743, 1725 (sh); δ_H (500 MHz, $CDCl_3$) 1.26 (s, 3H, αC**H**$_3$), 1.57 (s, 3H, βC**H**$_3$), 4.67 (s, 1H, **H**-3), 6.04 (d, *J* = 1.45 Hz, 1H, **H**-5), 6.31 (d, *J* = 1.45 Hz, 1H, sidechain C(O)C**H**=), 6.96 (s, 1H, C**H**$(Ar)_2$), 7.26-7.40 (m, 10H, Ar**H**). To a solution of this free acid (100mg, 0.236 mmol) in acetonitrile (5 mL) was added a solution of 7-hydroxy-4-methylcoumarin (62.5 mg, 0.355 mmoL) in acetonitrile (1mL), DMF (0.5 mL) and DMAP (14.4 mg, 0.118 mmol). The resulting solution was stirred at room temperature under nitrogen for 10 minutes. DCC (243 mg, 1.18 mmol) was added and the mixture was stirred for 75 min after which time TLC analysis (60:40 ethyl acetate/hexane) indicated the formation of one new product less polar than the starting acid. The reaction mixture was diluted with ethyl acetate (25 mL) and filtered by gravity. The filtrate was washed with water (3 x 30 mL). The ethyl acetate layer was dried, filtered and concentrated to give a brownish oil (291.5 mg). This was purified by column chromatography (60:40 ethyl acetate/ hexane) to give **A4** as a pale yellow solid (44 mg, 0.076 mmol, 32 %); mp 89 °C (dec); (Found C, 67.90; H, 4.95; N, 2.75%. $C_{33}H_{27}NO_7S$ requires C, 68.15; H, 4.68; N, 2.41%); ν_{max} (KBr)/cm^{-1} 1778 (lactam), 1735, 1709; δ_H (300 MHz, $CDCl_3$) 1.28 (s, 3H, αC**H**$_3$), 1.60 (s, 3H, βC**H**$_3$), 2.45 (s, 3H, C**H**$_3$Ar), 4.71 (s, 1H, **H**-3), 6.10 (s, 1H, **H**-5), 6.30 (s, 1H, C(O)C**H**=C, sidechain), 6.50 (s, 1H C**H**=C, coumarin), 6.96 (s, 1H, C**H**$(Ar)_2$), 7.16 (dd, *J* = 2.1, 8.7 Hz, 1H, Ar**H**, coumarin), 7.21 (d, *J* = 2.1 Hz, Ar**H**, coumarin), 7.29-7.39 (m, 10H, Ar**H**), 7.63 (d, *J* = 8.7 Hz, 1H, Ar**H**, coumarin); δ_C (75.47 MHz, $CDCl_3$) 18.76 (**C**H_3, coumarin), 25.30, 33.91 (α**C**H_3, β**C**H_3), 64.15, 69.15, 70.75 (**C**-2, **C**-3, **C**-5), 78.58 (**C**H$(Ar)_2$), 110.25, 114.42, 114.88, 117.66, 118.32 (C Ar coumarin, C(O)C=**C** sidechain, C=**C**(CH_3) coumarin), 125.56, 127.04, 127.56, 128.27, 128.46, 128.61, 128.67 (**C**, Ar), 138.95, 139.02 (C, Ar), 151.77, 152.23, 154.15, 159.36, 160.29, 161.47, 165.52, 166.52 (C, Ar coumarin, **C**-6, C(O)**C**=C coumarin), 4 × **C**=O).

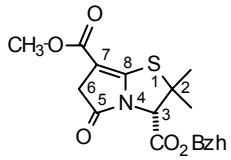
2-[1-(2-*t*-Butoxycarbonyl-1-methoxycarbonylethylene)]-5,5-dimethyl-4-diphenylmethoxycarbon yl-1,3-thiazolidine (B1).



A1 (140 mg, 0.29 mmol) was dissolved in methanol (5 mL) and to this was added triethylamine (150 μL, 1.076 mmol). The solution was stired at room temperature for 60 min at the end of which time TLC analysis showed the complete disappearance of **A1** and the appearance of one new product. Dichloromethane (20 ml) was added and the solution was extracted with water (2 x 20 mL), the organic layer was separated, dried and the solvent removed under reduced pressure to leave a yellow-green gel (97 mg). Purification by chromatography (silica gel, hexane/ethyl acetate 70/30) yielded **B1** as a pale yellow green glassy gel (70 mg, 0.137 mmol, 47%); (Found C, 65.28; H, 6.65; N, 2.70%. $C_{28}H_{33}NO_6S$

requires C, 65.73; H, 6.50; N, 2.74%); ν_{max} (KBr)/cm^{-1} 1744, 1728, 1708; δ_H (90 MHz, $CDCl_3$) 1.06 (s, 3H, αC**H**$_3$),[5] 1.38 (s, 9H, (C**H**$_3$)$_3$), 1.57 (s, 3H, βC**H**$_3$), 3.60 (br d, *J* = 11.4 Hz, N**H**, disappeared on addition of D_2O), 3.77 (s, 3H, C**H**$_3$O), 4.18 (d, *J* = 11.4 Hz, 1H, **H**-4, collapsed to a singlet on addition of D_2O), 6.06 (d, *J* = 9.0 Hz, 1H, **H**-2, collapsed to a singlet on addition of D_2O), 6.47 (s, 1H, sidechain C(O)C**H**=) 6.94 (s, 1H, C**H**(Ar)$_2$), 7.25-7.38 (m, 10H, Ar**H**); δ_C (75.47 MHz, $CDCl_3$) 26.86, 26.97 (α**C**H$_3$, β**C**H$_3$), 27.98 (C(**C**H$_3$)$_3$, 52.24 (**C**H$_3$O), 58.56, 62.38, 73.50, (**C**-5, **C**-4, **C**-2), 77.96 (**C**H(Ar)$_2$, 82.19 (O**C**(CH$_3$)$_3$, 126.91, 127.33, 127.85, 127.91, 128.34, 128.49, 128.59 (**C**, Ar) and ((O)CCH=**C**C(O)), 139.45, 139.54 (**C**, Ar), 141.70 ((O)C**C**H=CC(O)), 165.24, 166.54, 169.14 (3 × **C**=O).

Benzhydryl 7-methoxycarbonyl 2,2-dimethyl-5-oxo-2,3,5,6-tetrahydropyrrolo [2,1-*b*][1,3] thiazole-3-carboxylate (**C1**).

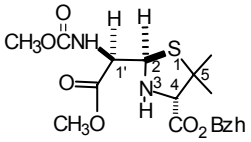


To a solution of **A2** (150 mg, 0.324 mmol) in methanol (5 mL) was added triethylamine (150 µl, 1.076 mmol) and the resulting solution was stirred for 35 min at room temperature at the end of which time TLC analysis showed the complete disappearance of **A2** and the appearance of one new product. Chloroform (20 ml) was added and the solution was extracted with water (2 x 50 mL), the organic layer was separated, dried and the solvent removed under reduced pressure to leave a brownish solid. Purification by chromatography (silica gel, hexane/ethyl acetate 50/50) yielded **C1** as a white solid (108 mg, 0.247 mmol, 76%); mp 54-56 °C (Found C, 65.22; H, 5.46; N, 2.95%. $C_{24}H_{23}NO_5S$ requires C, 65.89; H, 5.30; N, 3.20%); ν_{max} (KBr)/cm^{-1} 1736, 1690; λ_{max} (MeOH)/nm 218 (ε/M^{-1} cm^{-1} 18,182), 320 (ε 12,510); δ_H (300 MHz, $CDCl_3$) 1.30 (s, 3H, αC**H**$_3$), 1.71 (s, 3H, βC**H**$_3$), 3.51 (d, *J* = 23.11 Hz, 1H, -C**H**H-), 3.54 (d, *J* = 11.55 Hz, 1H, -CH**H**-), 3.75 (s, 3H, C**H**$_3$O), 4.46 (s, 1H, **H**-3), 6.92 (s, 1H, C**H**(Ar)$_2$), 7.25-7.38 (m, 10H, Ar**H**); δ_C (75.47 MHz, $CDCl_3$) 24.62 (α**C**H$_3$), 33.18 (β**C**H$_3$), 40.03 (**C**H$_2$, inverted in DEPT135), 51.34 (**C**H$_3$O), 62.66, 65.09 (**C**-2, **C**-3), 79.15 (**C**H(Ar)$_2$), 96.74 (**C**-7) 126.95, 127.57, 128.22, 128.48, 128.55, 128.58 (**C**, Ar), 138.78, 138.84 (**C**, Ar), 156.16 (**C**-8),[6] 163.99, 165.80, 172.54 (3 × **C**=O).

Benzhydryl 6-(methoxycarbonyl)aminopenicillanate (**E**).

To a solution of **1** (1.07 g, 2.80 mmol) in dichloromethane (53 mL) was added, methylchloroformate (324 µL, 4.19 mmol), pyridine (237 µL, 2.93 mmol) and DMAP (14 mg, 0.11 mmol). After stirring under nitrogen for 4.5 h at room temperature, the solution was washed with water (2 × 50 mL), 20% sodium chloride solution, separated and the organic layer was dried and removed under reduced pressure to leave a white solid (1.04 g) which was purified by chromatography (silica gel, hexane/ethyl acetate 60/40) to give **E** as a white solid (1.00 g, 2.27 mmmol, 81%); mp 132-133 °C (Found C, 62.43; H, 5.52; N, 6.26%. $C_{23}H_{24}N_2O_5S$ requires C, 62.71; H, 5.49; N, 6.36%); ν_{max} (KBr)/cm^{-1} 1800 (lactam), 1748, 1722; δ_H (300 MHz, $CDCl_3$) 1.27 (s, 3H, αC**H**$_3$), 1.62 (s, 3H, βC**H**$_3$), 3.72 (s, 3H, C**H**$_3$O), 4.54 (s, 1H, **H**-3), 5.43-5.57 (m, 2H, **H**-6, N**H**) overlapping with 5.56 (d, *J* = 3.9 Hz, 1H, **H**-5), 6.94 (s, 1H, C**H**(Ar)$_2$), 7.30-7.37 (m, 10H, Ar**H**); δ_C (75.47 MHz, $CDCl_3$) 26.67, 32.19 (α and β**C**H$_3$), 52.92 (**C**H$_3$O), 60.67, 65.07, 68.31, 70.43 (**C**-2, **C**-3, **C**-5, **C**-6), 78.44 (**C**H(Ar)$_2$), 126.98, 127.57, 128.24, 128.46, 128.61, 128.67 (**C**, Ar), 138.97, 139.03 (**C**, Ar), 155.58, 166.76, 173.99 (3 × **C**=O)

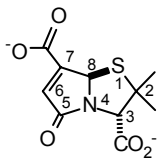
2-[1-(methoxycarbonyl)aminomethyl-1-methoxycarbonyl]-5,5dimethyl-4-diphenylmethoxy carbonyl-1,3-thiazolidine (F).



To a solution of **E** (149.5 mg, 0.34 mmol) in methanol (5 mL) was added triethylamine (150 µL, 1.076 mmol) and the resulting solution was stirred for 40 min at room temperature after which time TLC analysis indicated complete conversion of **E** to one new product. Dichloromethane (20 mL) was added,

the solution was extraced with water (2 × 50 mL), the organic layer was separated, dried and removed under reduced pressure to leave a white solid (129 mg) which was purified by chromatography (silica gel, ethyl acetatehexane 40/60) to leave **F** as a white solid (94 mg, 59%); mp 51-52 °C (Found C, 60.70; H, 6.04; N, 5.75%. $C_{24}H_{28}N_2O_6S$ requires C, 61.00; H, 5.97; N, 5.93%); ν_{max} (KBr)/cm^{-1} 1733 (br); δ_H (300 MHz, $CDCl_3$) 0.99 (s, 3H, αC**H**$_3$), 1.57 (s, 3H, βC**H**$_3$), 2.2 (br baseline peak, thiazolidine N**H**), 3.70 (s, 3H, carbamate C**H**$_3$O), 3.76 (s, 3H, ester C**H**$_3$O), 3.85 (s, 1H, **H**-4) 4.35 (br s, 1H, **H**-1'), 5.08 (d, *J* = 4.2 Hz, **H**-2), 5.46 (br s, 1H, carbamate N**H**), 6.95 (s, 1H, C**H**(Ar)$_2$), 7.27-7.40 (m, 10H, Ar**H**); δ_C (75.47 MHz, $CDCl_3$) 26.42, 26.93 (α and β**C**H$_3$), 52.58, 52.68 (carbamate **C**H$_3$O and ester **C**H$_3$O), 59.04 (**C**-1'; enhanced in DEPT90), 59.11 (**C**-5), 66.41, 72.89 (**C**-2, **C**-4), 78.30 (**C**H(Ar)$_2$), 126.87, 127.81, 128.14, 128.48, 128.60, 128.64 (**C**, Ar), 139.13, 139.22 (**C**, Ar), 156.9 168.66, 170.54 (3 × **C**=O).

Kinetic analysis of A3' and isolation of hydrolysis co-product C' (as the free acid).



A3' (40 mg, 0.136 mmol) was dissolved in D_2O buffer (1.0 mL, 0.2 M phosphate, pD 7.2). To this solution was added β-lactamase I ex *B. cereus* (Sigma) (25 mg) and reaction progress was monitored by recording the ^{1}H NMR (90 MHz) spectrum as a function of time. On completion of the hydrolysis distilled water (20 mL) was added, the solution was acidified to pH 2.2 and extracted with ethyl acetate (2 × 20 mL). The organic layer was dried and concentrated under reduced pressure to leave a light-yellow solid (30 mg); δ_H (300 MHz, D_2O buffer, pD 7.2) 1.60 (s, 6 H, α and βC**H**$_3$), 4.20 (s, 1H, **H**-3), 6.01 (s, 1H, **H**-8), 6.49 (s, 1H, **H**-6); δ_C (75.47 MHz, D_2O buffer, pD 7.2) 27.95, 31.72 (α and β**C**H$_3$), 63.71, 70.24, 72.08 (**C**-2, **C**-3, **C**-8), 129.55 (**C**=**C**), 161.59, 176.78, 176.92 (3 × **C**=O); ESI-HRMS (MeOH) *m*/*z* [M - H]$^-$ calcd for $C_{10}H_{11}NO_5S$ 256.0285, found 256.0289.

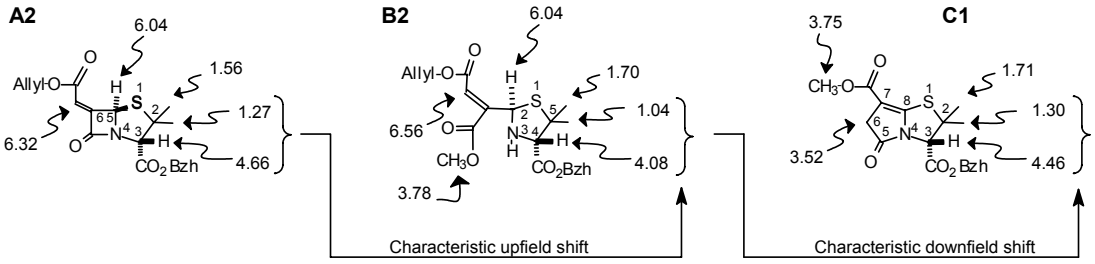


Chart-S1. Summary of characteristic ^{1}H NMR chemical shift patterns of the benzhydryl esters **A2**, **B2** and **C1**.

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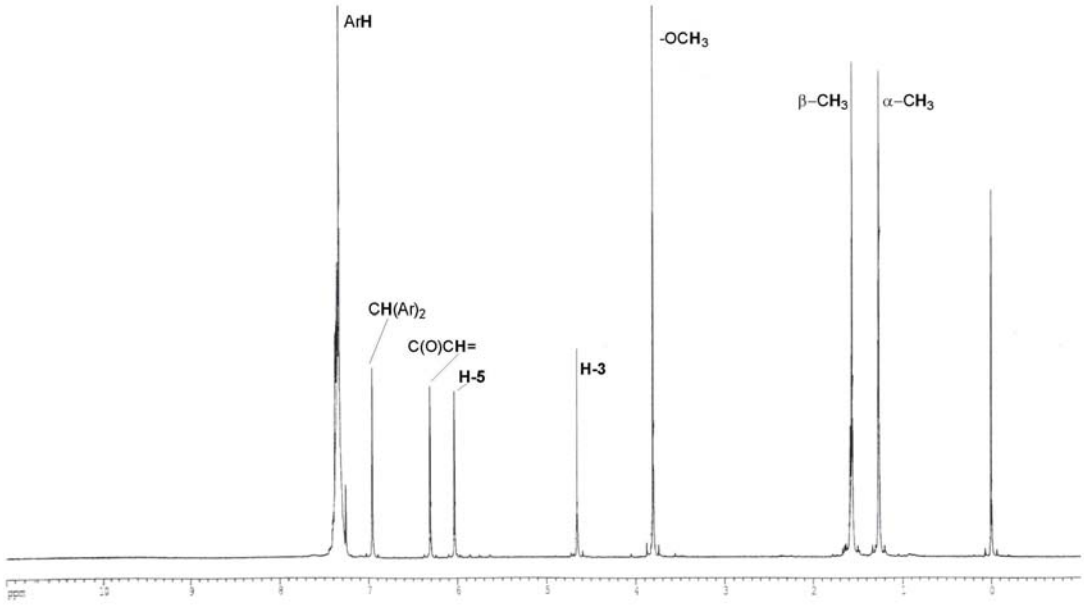

Figure S1; ^{1}H NMR spectrum of **A3**

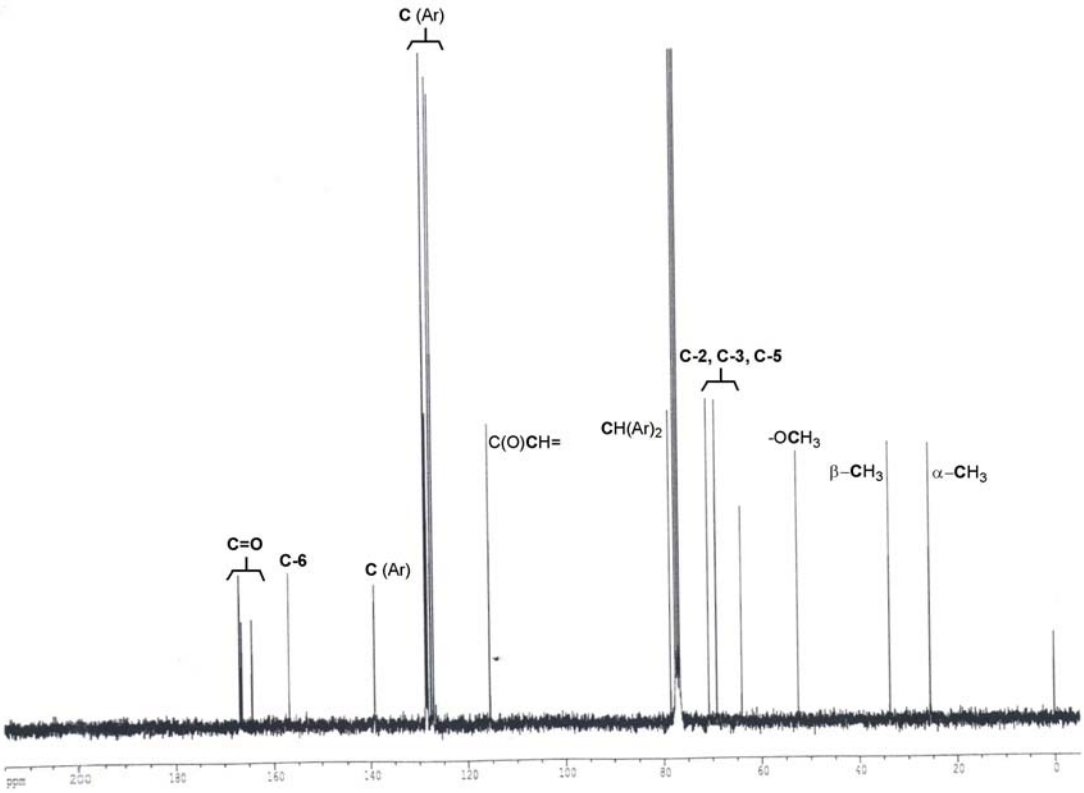

Figure S2; ^{13}C NMR spectrum of **A3**

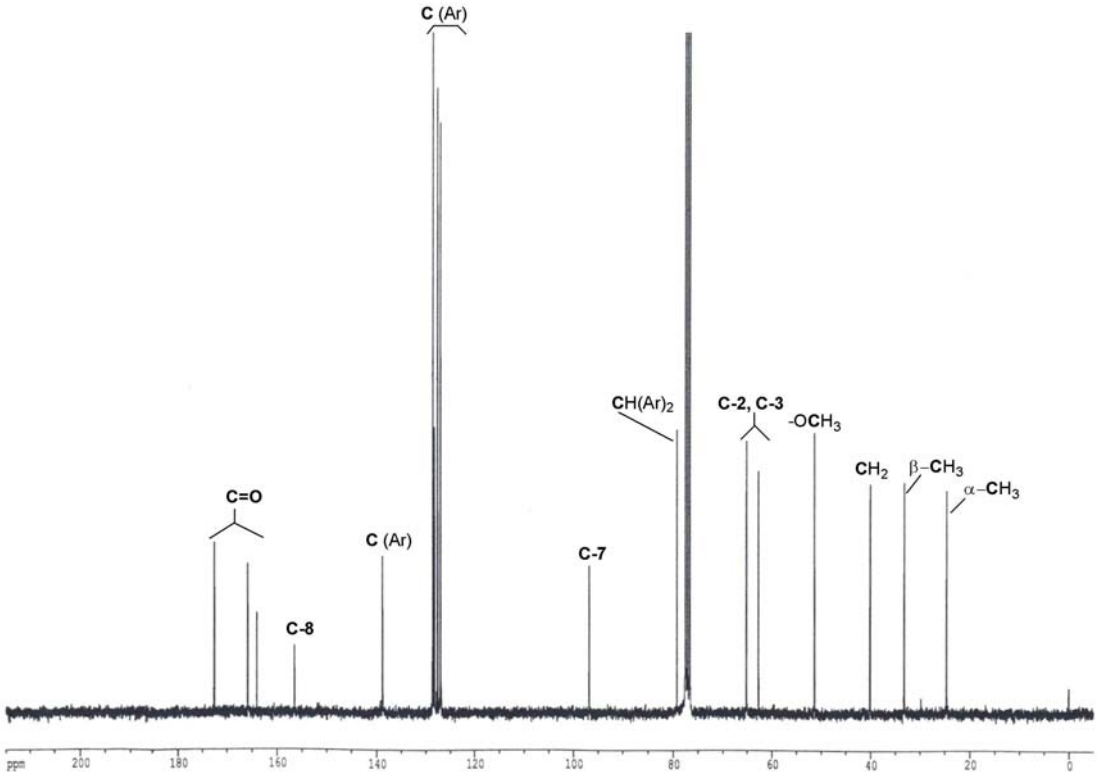

Figure S3; ^{13}C NMR spectrum of **C1**

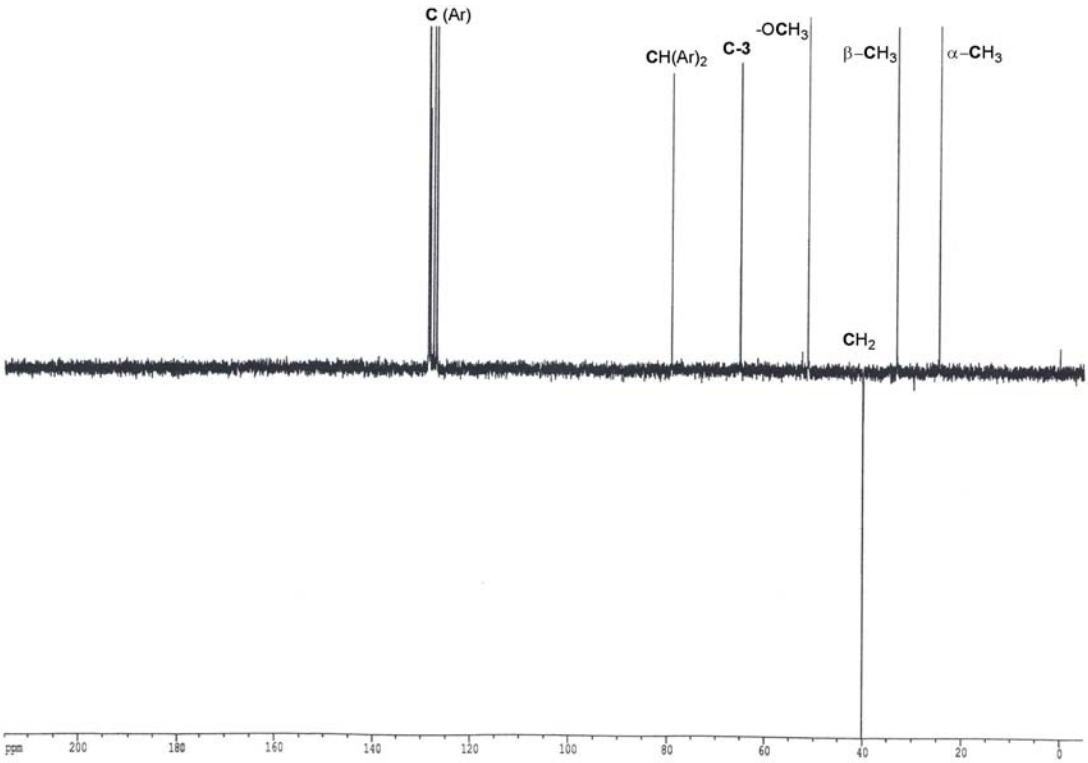

Figure S4; ^{13}C DEPT 135 of **C1**

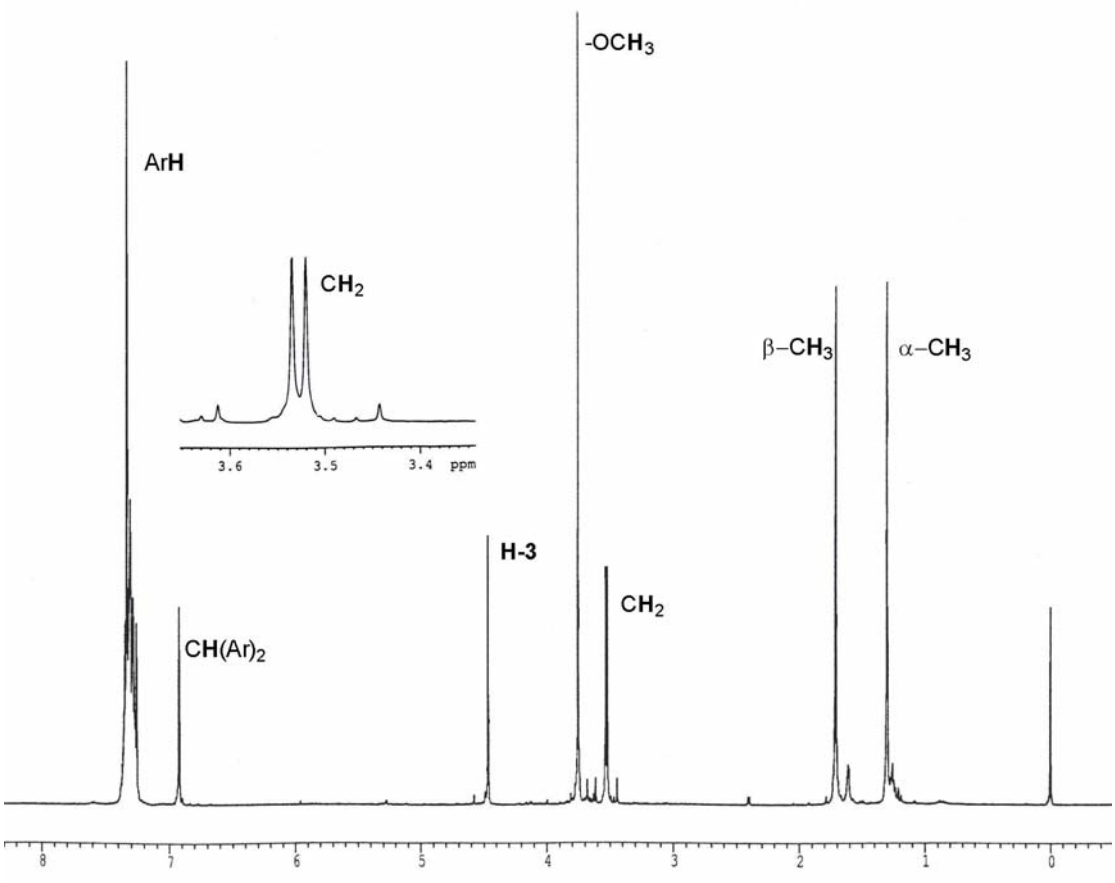

Figure S5; ^{1}H NMR spectrum of **C1**