“Synthesis of a bicyclic oxo-γ-lactam from a simple caprolactam derivative”

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Abstract

Synthesis of the 6-azabicyclo[3.2.1]octane ring system, *via* Dieckmann cyclization, is described. Ring closure involves reaction of a caprolactam enolate with a C6 ester, the reactive axial conformation of which is promoted by the presence of an *N*-butyloxycarbonyl group on the lactam nitrogen. The results will enable the synthesis of new bridged caprolactams for testing as antibacterials and nucleophilic enzyme inhibitors.

The β-lactams remain the most important antibacterials; they work by reaction with a nucleophile serine residue in penicillin binding proteins (PBPs), which catalyse essential transpeptidase reactions during bacterial cell-wall peptidoglycan biosynthesis. A common mechanism of resistance to the β-lactam antibacterials, involves β-lactamases, which catalyse β-lactam hydrolysis. All clinically used PBP inhibitors are β-lactams and until recently this has been the case for β-lactamase inhibitors.[1] Following on from synthetic γ-lactam analogues of the β-lactams and the discovery of the natural product lactivicin, the cyclic urea avibactam has recently been introduced as a broad spectrum serine β-lactamase inhibitor.[2] However, while the β-lactam based inhibitors react irreversibly with the nucleophile serine of the PBPs and β-lactamases, avibactam reacts reversibly with its target serine β-lactamases.[3] The discovery of avibactam has stimulated interest in non β-lactam inhibitors of the serine β-lactamases and PBPs. We have been interested in bridged lactams as inhibitors of nucleophilic serine / threonine / cysteine enzymes; however, for ring sizes > 6 there are only limited reports on their synthesis. Here we describe the synthesis of the 6-azabicyclo[3.2.1]octane bridged ring system, starting from a readily available caprolactam precursor (Fig. 1).

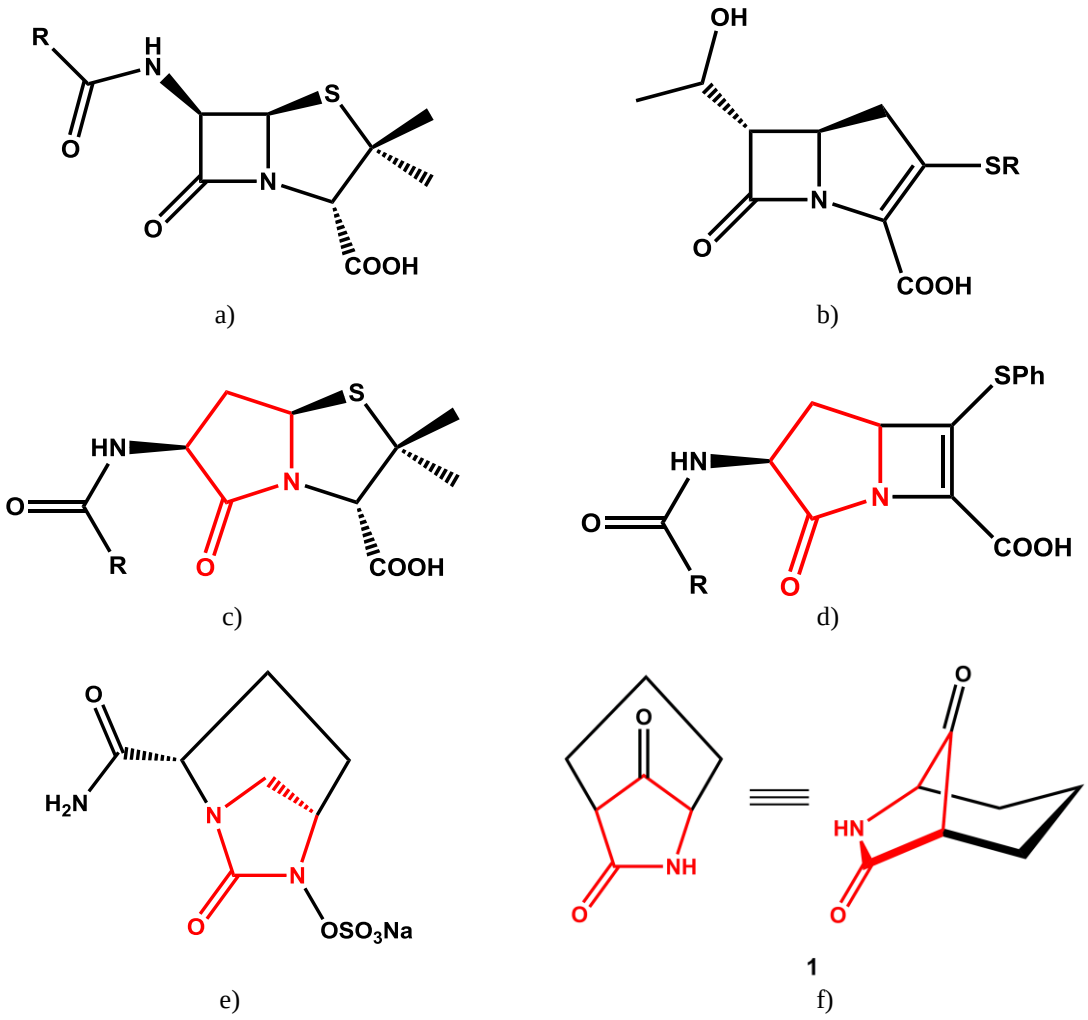

Fig. 1 Examples of β-lactam antibacterials, β-lactam inhibitors and non-β-lactam analogues: a) penicillins, b) carbapenems, c) an inactive γ-lactam analogue, d) an active γ-lactam analogue, e) avibactam and f) the target of the current work (**1**) which has a 6-azabicyclo[3.2.1]octane core ring system.

The 6-azabicyclo[3.2.1]octane ring system is present in a wide range of biologically active compounds and is isomeric with the tropane nucleus present in alkaloids, including cocaine and atropine.[4] Preparation of respective 6-azabicyclo[3.2.1]octane derivatives and related

compounds is restricted to the intramolecular ring closure of γ-lactam derivatives,[5] amide formation in substituted cyclohexanes[6], and Diels-Alder reaction of appropriately unsaturated γ-lactams with acrylic acid.[7] There is only one reported route to a 7,8-dioxo-6-azabicyclo[3.2.1]octane derivative of (**1**), which employs semipinacol rearrangement of a β-lactam precursor.[8] We envisaged bicycle **1** could be succinctly prepared *via* cyclization of a simple caprolactam *via* Dieckmann cyclization. We anticipated that then Dieckmann cyclization may only proceed efficiently, when the ester group adopts an axial position (Fig. 2a/b/c).

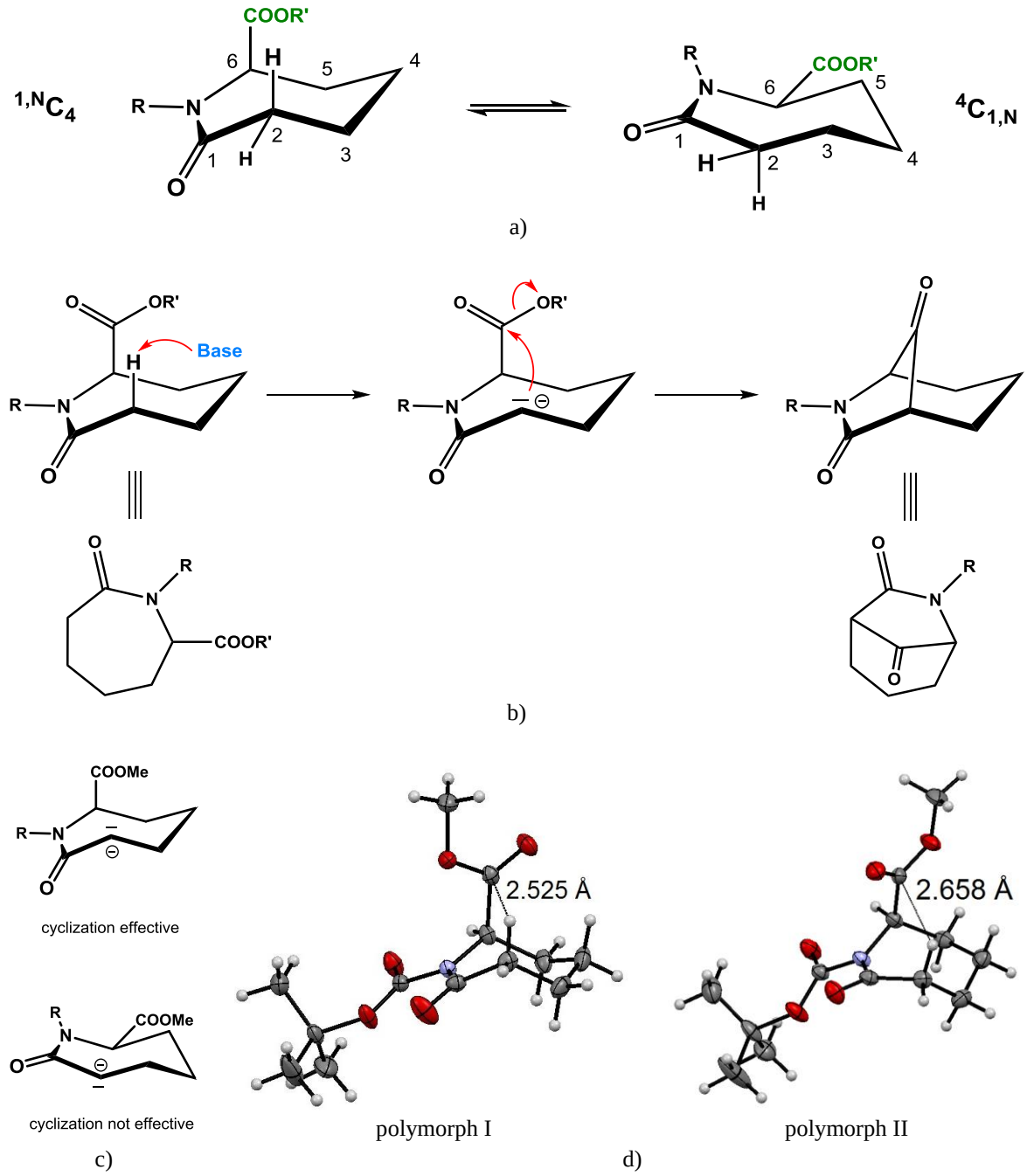

Fig. 2 Proposed Dieckmann cyclisation to give the 6-azabicyclo[3.2.1]octane ring system. a) The two energetically favoured 'pseudo chair' conformations of an *N*-substituted caprolactam methylester. b) Synthesis of **1** *via* Dieckmann cyclization. c) Only the axially positioned ester group can react *via* the desired Dieckmann cyclization. d) View from a crystal structure of **3** showing that the –COOMe group adopts an axial conformation, as observed for both polymorphs. In solution an equilibrium between the 'axial' and 'equatorial' conformers (70 % : 30 %) is observed.[12]

Caprolactams can adopt (pseudo) chair, boat or transition (twist boat) forms.[9] In the 'chair' form, two energetically favoured conformations are manifested ($^{1,N}C_4$ and $^{4}C_{1,N}$) assuming an planar amide.[10] Similar to cyclohexane chair conformations, caprolactams feature axial and equatorial positions of ring hydrogens and respective substituents. The axial substituents are higher in energy than equatorial ones (as shown for C-2/C-6 substituted caprolactams).[11] In a previous study,[12] we investigated the influence of a second substituent on the conformation of the C-6 caprolactam methylester (**2**) (Fig. 3). Importantly for the current purpose, we observed by crystallography that introduction of the bulky *tert*-butyloxycarbonyl (Boc) group at the caprolactam nitrogen causes the C-6 ester to adopt the normally energetically disfavored axial conformation. Note the short distance of 2.53 Å (polymorph I)[12] and 2.66 Å (polymorph II, Tabs. S1 and S2, Scheme S1, ESI), respectively, between the C-6 ester carbonyl and the axial C-2 hydrogen as observed by X-ray crystallography (Fig. 2d). Indeed, treatment of **3** with LiHDMS (lithium bis(trimethylsilyl)amide) produced the desired bicyclic lactam in its protected form (**1a**) in low (8 %) yield (Fig. 3). Although it is likely that the cyclisation yield can be optimised, initial attempts to improve the yield by use of other bases (DBU, 1,8-diazabicyclo[5.4.0]undec-7-ene, and potassium *tert*-butoxide) and varying temperature (including use of LiHMDS at room temperature), and, somewhat suprisingly, concentration[13] (Tab. S3, ESI) were unsuccessful.

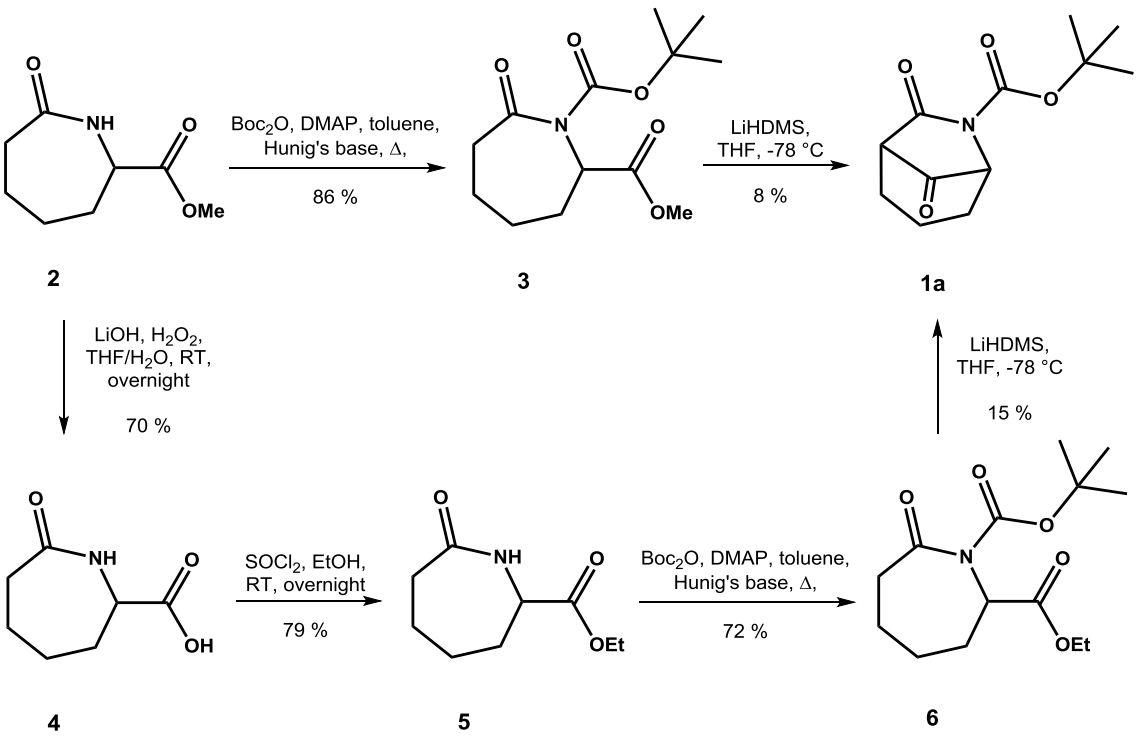

Fig. 3 Synthetic pathway to bicycle **1a**.

Caprolactam **3** features two C-H acidic position (C-2 and C-6) and a competing deprotonation may be one reason for the low yield of the bicyclic lactam **1a** in the cyclisation step. To investigate the proposal of competing C-6 deprotonation, Boc-protected ethylester **5** was prepared. Thus, caprolactam methylester **2** was saponified to yield free acid **4**, which was esterified with EtOH and, subsequently, treated with Boc anhydride. When **5** was subjected the Dieckmann cyclization, a higher yield of **1a** (15 %) was achieved (Fig. 3), likely due to reduced C-6 deprotonation relative to **3** due to the higher steric demand of the ethyl ester.

^{1}H and ^{13}C NMR (Fig. S1, ESI) as well as COSY analyses (Fig. S2, ESI) support the assigned structure of **1a.** Of note, the ^{1}H spectrum exhibits a ‘doublet of doublets of doublets‘ (ddd) coupling for *H2* and *H6*. The third coupling likely results from $^{4}J_{H,H}$ long-range ‘W’ coupling[14,15] of *H2* with *H6* over the keto bridge. For the bicyclic lactam **1a**, *H2* and *H6* couple with ^{4}J values of 4.89 and ^{4}J=4.88 Hz respectively; the analogous value for cyclobutanone is 4.8 Hz.[16] By contrast the ^{3}J-couplings are rather low, both for the coupling of *H2* with *H3*/*H3’* (^{3}J=2.50/1.95 Hz) and of *H2* with *H6* (^{3}J=1.88/1.33 Hz) (Fig. 4; Fig. S3, ESI).

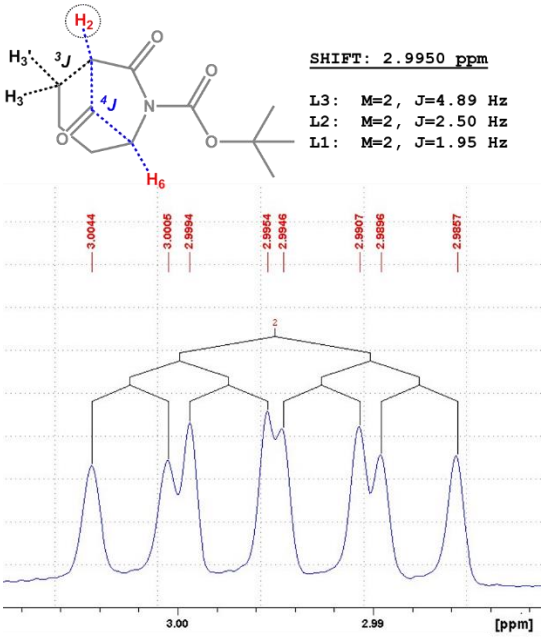

Fig. 4 Close up view from the ^{1}H NMR (125 MHz) spectrum of **1a**; the *H2* signal with the respective ddd coupling pattern and the respective $^{3}J_{H,H}$ and $^{4}J_{H,H}$ values is shown.

Bicycle **1a** crystallized from ethyl acetate and cyclohexane as its corresponding hydrate **1b** (Fig. 5a), a phenomen which was already observed with a related compound.[4] The plate like twinned crystals are in spacegroup *P*-1 with molecules featuring (*R*,*R*) and (*S*,*S*) stereochemistry. As expected, the five-membered ring of **1b** adopts an envelope conformation, while the caprolactam adopts the rarer boat conformation (Fig. 5b). A comprehensive comparison of bond lengths and angles with related compounds can be found in the Supplementary Material (Tab. S4, ESI).The molecules of **1b** arrange in hydrogen bonded ribbons running in the direction of the crystallographic *a* axis. In the ribbons two diols face each other making an $R_2^2(8)$ motif and these dimers then bond into C6 chains *via* the ring carbonyl groups (Fig. 5c; Tab. S5, ESI). The ribbons stack up on each other with only weak C-H···O(-H) hydrogen bonds in the direction of the crystallographic *b* axis. The *tert*-butyl groups point outward from the sheet assembled *via* these interactions and stacking errors of these sheets cause the crystals to be twinned.

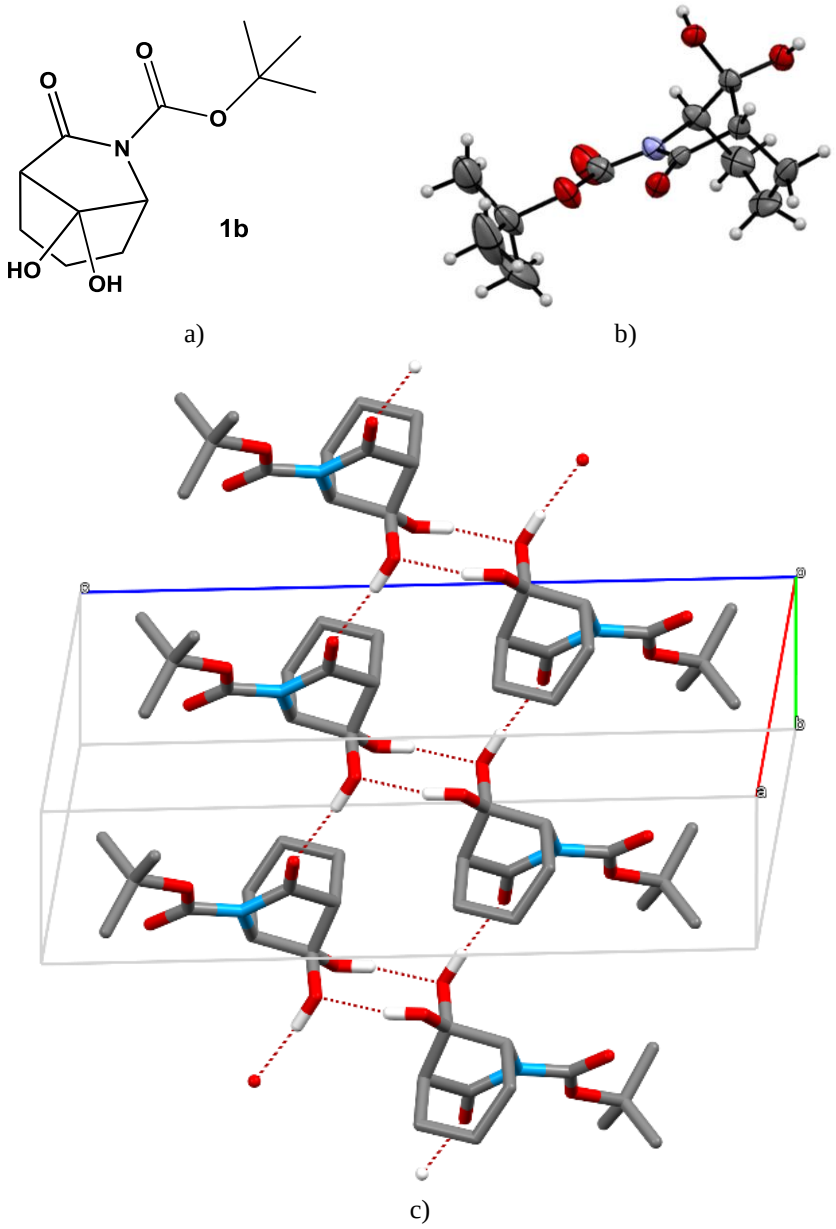

Fig. 5 a) Molecular formula of the hydrate of **1a**, *i.e.* **1b**. b) Ortep plot at 50 % probability of crystal structure of **1b**. c) Hydrogen bonded ribbons of **1b** running along the crystallographic *a* axis.

Conclusions

Overall, we have described the concise synthesis of a bridged caprolactam ring system, *via* Dieckmann cyclization. This route builds upon work that has defined the transformations of readily accessible caprolactam derivatives. Closure to give the 6-azabicyclo[3.2.1]octane ring system involves the reaction of a caprolactam enolate with an C-6 ester in an axial conformation. The presence of the reactive axial conformation is promoted by an *N*-butyloxycarbonyl group on the caprolactam nitrogen. The yield is increased on with use of an ethyl, rather than a methyl, ester, likely due to diminished C-6 deprotonation with the ethyl ester. Future work can focus on a deeper understanding of the mechanism and minimising side reactions. The results will enable the synthesis of 6-azabicyclo[3.2.1]octane ring derivatives for testing as antibacterials and nucleophilic enzyme inhibitors.

Experimental

Materials and Methods

Melting points were determined using a microscope heating stage PHMK Rapido (VEB Dresden Analytik). IR spectra were measured using a Bruker Tensor 27 ATR-FT-IR with the ATR method. NMR spectra were recorded using a Bruker Avance DRX 500 spectrometer at 500.13 MHz (^{1}H-NMR) and 125.77 MHz (^{13}C-NMR), respectively. Chemical shifts δ are reported in parts per million relative to the internal reference TMS. Multiplicity is abbreviated as follows: s (singlet), d (doublet), t (triplet) and m (multiplet). Mass spectra were measured on a Varian 320 MS. Elemental analyses were performed on a Heraeus CHN-Rapid Analyzer.
All reactions involving moisture sensitive reagents were carried out under a nitrogen atmosphere. Cooling was performed in ice-water baths (0 °C) or dry ice-acetone baths (-78 °C). Anhydrous solvents were used as supplied. Thin layer chromatography (TLC) was performed on Merck DC-Kieselgel 60 F 254 0.2 mm precoated plates with fluorescence indicator. Visualization of spots was achieved using UV light (254 nm) and by developing in a basic solution of $KMnO_4$ followed by heating.
The syntheses of the starting caprolactam **2**, the Boc-protected lactam **3** and the free acid **4** have been described previously.[12] Synthetic procedures for compounds **5** and **6** are given in the Supplementary Material.

***tert*-Butyl 7,8-dioxo-6-azabicyclo[3.2.1]octane-6-carboxylate (1a).** To a stirred solution of the appropriate Boc-protected lactam (**3** or **6**, resp.) (1 eq.) in dry THF (15 ml) at -78 °C was added a 1M solution of lithium hexamethyldisilazide (LiHDMS) in ethylbenzene/THF (1.1 eq.). The reaction mixture was then stirred at -78 °C for 3 h. Subsequently, the reaction was quenched with sat. aqueous ammonium chloride solution (30 ml) at -78 °C and extracted with ethyl acetate (3x). The combined organic phases were dried (Na_2SO_4), then filtered. Evaporation of the solvent yielded a dark, oily residue which was separated by flash column chromatography (SiO_2; *n*-hexane/ethyl acetate = 1:1 → ethyl acetate) to yield 8 % (43 mg, 0.18 mmol) (methyl ester) or 15 % (22 mg, 0.092 mmol) (ethyl ester), respectively, of a white solid. Mp. 93-94 °C. R_f = 0.30 (SiO_2; *n*-hexane/ethyl acetate = 1:1). ^{13}C NMR (100 MHz, $CDCl_3$): δ = 207.3 (*C*O), 168.9 (*C*ONH), 148.4 (N*C*OO^{t}Bu), 84.1 (CH_3)$_3$, 64.9 (CO*C*HCO), 55.0 (N*C*HCO), 33.2 (*C*H$_2$), 32.7 (*C*H$_2$), 28.0 (CH_3)$_3$, 17.1 (*C*H$_2$). ^{1}H NMR (400 MHz, $CDCl_3$): δ = 4.39 (m, 1H, NHC*H*), 2.99 (m, 1H, COC*H*CO), 2.44-2.31 (m, 2H, C*H*$_2$), 2.05-1.96 (m, 1H, C*H*$_2$), 1.95-1.88 (m, 1H, C*H*$_2$), 1.84-1.75 (m, 2H, C*H*$_2$), 1.54 (s, 9H, C(C*H*$_3$)$_3$). IR: 3391, 2992, 2932, 2874, 1769, 1752, 1713, 1448, 1393, 1365, 1326, 1305, 1249, 1220, 1154, 1089, 1068, 1052, 1014, 993, 975, 953, 888, 865, 712. m/z = 238.11 [M-H$^+$], calc. 238.12.

X-ray crystallography

Bicycle **1a** was crystallized from ethyl acetate and cyclohexane using the vapor diffusion approach resulting in hydrate **1b**. Crystals suitable for single crystal X-ray diffraction studies of polymorph II of compound **3** were obtained by crystallization from ethyl acetate/*n*-hexane (1:2) and have a melting point of 69-71 °C; this is about 20 K higher than observed for polymorph I.
Single crystal X-ray diffraction was performed at 173K with a Bruker D8 Venture diffractometer using a Cu-K$_\alpha$ source. Structure solution was carried out with shelxt[17] and structure refinement with shelxl[18] was finished using ShelXle[19] software. The twin matrix for **1b** was acquired from twinrotmat in Platon.[20] For crystal data and refinement parameters see ESI. CCDC numbers 1528320 (**1b**) and 1522636 (**3**).

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