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Synthesis of modular dipeptide mimetics on the basis of diazabicycloalkanes and derivatives thereof with sulphur containing side chains

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Summary. We present the synthesis of new modular dipeptide mimetics based on diazabicycloalkane backbones. These diazabicycloalkanes are ligands for the prostate specific membrane antigen (PSMA), a well known tumor marker. Our previously described synthetic route to enantiomerically pure diazabicycloalkanes is extended to yield polyfunctional diazabicycloalkanes with a modular character using a new ring closing methodology. This, finally, allows us to attach linker moieties to different positions of the diazabicycloalkane scaffold providing conjugation sites to other functional molecules such as markers or cytostatic compounds. Furthermore, successful synthesis of sulphur-containing dipeptide analogues as for example $CysX_{AA}$- or $HCysX_{AA}$-mimetics on the basis of diazabicycloalkanes is described.

Keywords: Prostate cancer – PSMA – Diazabicycloalkanes – Stereoselective synthesis

Introduction

Prostate cancer accounts for 13 per cent of all new cancer diagnoses and nearly nine per cent of all cancer deaths. These dramatically high rates are due to mainly two factors. On the one hand, traditional chemotherapies, typically based on targeting rapidly proliferating cells, turn out to be ineffective due to very low proliferation rates of androgen independent prostate cancer cells (Jayne Oliver et al., 2003). On the other hand there is still a lack of reliable diagnostic tools to allow secure detection of malignant prostate cancer cells at early stages of the disease, at which hormonal treatment is promising. Therefore, there is an urgent need to develop a fast and reliable diagnostic tool for prostate cancer. An appropriate target for the treatment and diagnosis of prostate cancer is the prostate specific membrane antigen (PSMA) (see for example Harada et al., 2003).

We have reported the synthesis of diazabicycloalkanes of type **1** and demonstrated their ability to bind PSMA with low micromolar affinities (A number of our previously described dipeptide mimetics (Maison et al., 2004) proofed to be ligands for PSMA in an *in vitro* assay (Slusher et al., 1990). While rigid azabicycloalkanes of type **2** were known and had been used to probe conformation-activity relationships (Hanessian et al., 1997) and a whole set of structurally related bicyclic analogues were prepared and incorporated into peptides as turn mimetics (Belvisi et al., 2001; Grossmith et al., 1999; Halab et al., 2000), only few conformationally restricted analogues **1** on the basis of a diazabicycloalkane scaffold were described (St-Denis et al., 1998; Chan et al., 1999; Tong et al., 2000). We developed recently a short and efficient synthetic route to diazabicycloalkanes of type **1** (Fig. 1) (Maison et al., 2002, 2004). However, two drawbacks limited the versatility of this route: first, linker moieties allowing conjugation to other functional molecules, such as fluorescence markers for *in vivo* imaging, could not be introduced to any position of the scaffold but to *N*-4 *via* acylation. This still allows the application of diazabicycloalkanes as PSMA ligands but a potential use of the diazabicycloalkane scaffold in combinatorial chemistry is restricted. In addition, sulphur containing amino acids such as cysteine or methionine cannot be introduced in diazabicycloalkane peptide mimetics. This is due to an oxidative cleavage as a key step in our sequence. Since thiols and thioethers are susceptible to oxidation, the choice of the *N*-terminal amino acid was restricted, excluding cysteine and methionine.

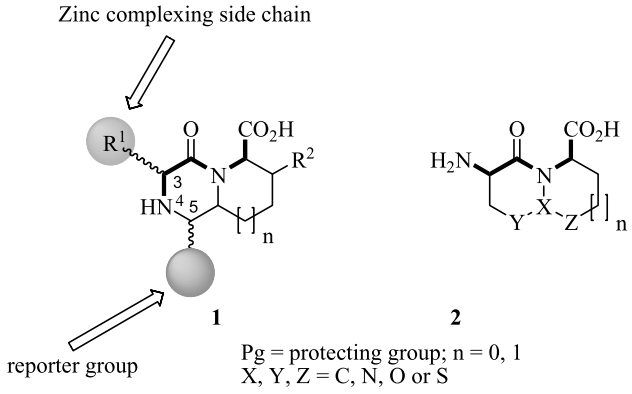


Fig. 1. General structures of diazabicycloalkanes **1** and known turn mimetics **2**

We describe herein an improved synthetic procedure, that addresses drawbacks in our previous synthetic scheme and allows introduction of linker moieties at different positions of the diazabicycloalkane scaffold **1**. Furthermore, introduction of sulphur containing amino acid side chains has been completed, providing access to $CysX_{AA}$ and *H*$CysX_{AA}$ dipeptide mimetics. Such mimetics are particularly interesting because PSMA is a zinc protease and a number of ligands with Zn^{2+} complexing side chains have been shown to bind PSMA with nanomolar affinities (Majer et al., 2003).

Material and methods

General remarks

If indicated with 'dry' solvents were purified prior to use as follows: methanol was distilled from magnesium, dichloromethane was distilled from $CaCl_2$, THF and Et_2O were distilled from sodium and benzophenone under nitrogen. – Thin layer chromatography (TLC) analyses were performed on silica gel 60 F_{254} plates from Merck. Spots were visualized with a Mo-Ce staining mixture (50 g ammonium molybdate-(VI)-tetrahydrate, 1 g cerium-(IV)-sulfate in 100 ml concentrated H_2SO_4 and 900 ml water). – For preparative chromatography Merck silica gel 60, 230–400 mesh was used. – Optical rotations $[\alpha]^{20}_D$ were determined with a Perkin Elmer polarimeter (241 MC) in $CHCl_3$ at 20°C, c = 1 unless otherwise indicated. Specific rotations are based on the equation $[\alpha] = (100 \cdot \alpha / 1 \cdot c)$ and are reported as unitless numbers where c is in g/100 mL and l is in decimeters. The units $[\alpha]$, $(\deg \cdot mL)/(g \cdot dm)$, are implicit and are not included with the represented value. – ^{1}H-NMR and ^{13}C-NMR spectra were recorded with a Bruker-Karlsruhe AMX 400 spectrometer (400 MHz/100.6 MHz) or on a Bruker-Karlsruhe DRX 5001 spectrometer (500 MHz/125.8 MHz). Chemical shifts δ, are presented in part per million (ppm) and coupling constants *J*, in Hertz (Hz) from tetramethylsilane (TMS, 0 ppm) as the internal standard for $CDCl_3$ and residual solvent peaks for [D_4] MeOH. – Mass spectra were obtained with a VG/70–250 F (VG Analytical) instrument in FAB mode in a *p*-nitrobenzylalkohol matrix or on a MAT 95 Trap XL (Thermo Finnigan) instrument in ESI mode (positive mode) using polypropylene glycole or polyethylene glycole as internal standard. – The following compounds were synthesized according to literature procedures or previously described protocols: *tert*-butyl (triphenylphosphanylidene) acetate (Cooke and Burman 1982), 6-(*tert*-butoxycarbonylamino)hexanoic acid (Klyashchitskii et al., 1981) and **18** (Maison et al., 2004) as well as **8b**, **8c**, **8e**, **10b**, **10c**, **10e** and the TBDMS-protected precursor of **19e** (Maison et al., 2004) (*tert*-butyl-((3*S*,6*S*,7*S*,9*S*)-2-benzoyl-7-(2-*tert*-butoxycarbonyl-ethyl)-3-(*tert*-butyldimethyl-silyloxymethyl)-4-oxooctahydropyrrolo[1,2-*a*]pyrazin)-6-carboxylate). EA = ethyl acetate; PE = petroleum ether; 50/70 = boiling range 50–70°C.

General procedure for the synthesis of dipeptides **8a** *and* **8d** *(GP 1)*

tert-Butyl-(1*S*,3*S*,4*R*)-2-[(1*R*)-phenylethyl]-2-aza-bicyclo[2.2.1]hept-5-ene-3-carboxylate (**18**) (3.28 g, 14.3 mmol) and the appropriate amino acid (16.3 mmol) were dissolved in 110 mL of dry DMF. A solution of HOBt (2.01 g, 14.3 mmol) in 40 mL of dry DMF was added and the solution was cooled to 0°C. DCC (3.56 g, 17.2 mmol) was added and the resulting suspension was warmed to rt and stirred under nitrogen for 12 h. DCU was filtered off and DMF was removed *in vacuo*. The crude product was purified by column chromatography on silica gel to give dipeptides **8** as colorless sticky solids.

Dipeptide **8a**

The title compound was synthesized according to GP 1 from Cbz-Asp(OMe)-OH (4.58 g, 16.3 mmol) and was obtained in 4.56 g (9.26 mmol, 65%) yield. $R_f = 0.31$ (EA/PE, 7:3; Mo/Ce). $[\alpha]^{20}_D = -56.1$ (c = 1.0, $CHCl_3$). ^{1}H NMR (500 MHz, [D_4] MeOH, 8:2 mixture of rotamers): $\delta = 7.27$–7.36 (m, 5H), 5.01–5.13 (m, 2H), 4.86 (dd, 0.8H, $J = 6.3$, 7.9 Hz), 4.41 (dd, 0.2H, $J = 4.4$, 9.8 Hz), 4.31 (br, 0.2H), 4.24 (br, 0.8H), 4.02 (d, 0.8H, $J = 4.7$ Hz), 3.87 (d, 0.8H, $J = 5.3$ Hz), 3.71 (s, 0.8H), 3.68 (s, 2.4H), 3.65 (s, 0.6H), 3.36–3.54 (m, 0.6H), 2.86 (dd, 0.2H, $J = 6.9$, 16.7 Hz), 2.82 (dd, 0.8H, $J = 6.0$, 16.1 Hz), 2.65–2.70 (m, 0.2H), 2.65 (dd, 0.8H, $J = 8.5$, 16.4 Hz), 2.60 (br, 0.2H), 2.46 (br, 0.8H), 1.84–1.98 (m, 2H), 1.44 (s, 7.2H), 1.43 (s, 1.8H). ^{13}C NMR (100 MHz, [D_4] MeOH, mixture of rotamers): $\delta = 172.2$, 170.8, 170.1, 138.0, 129.0, 129.4, 129.0, 128.8, 83.9, 83.0, 73.5, 73.2, 67.9, 62.9, 62.6, 62.1, 61.3, 52.5, 52.4, 50.8, 50.5, 50.0, 48.2, 38.5, 37.3, 29.8, 28.2, 28.2. HRMS (FAB) calcd. for $C_{24}H_{33}N_2O_9$ (MH^+) 493.2186, found 493.2189.

Dipeptide **8d**

The title compound was synthesized according to GP 1 from Cbz-HSer(OTBDMS)-OH (8.08 g, 22.0 mmol) and was obtained in 6.60 g (11.4 mmol, 59%) yield. $R_f = 0.18$ (EA/PE, 1:2; Mo/Ce). $[\alpha]^{20}_D = -27.2$ (c = 1.0, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 7.28$–7.38 (m, 5H), 5.73 (d, 1H, $J = 8.9$ Hz), 4.99–5.13 (m, 2H), 4.45–4.69 (m, 1H), 4.30 (s, 1H), 4.06 (d, 1H, $J = 5.7$ Hz), 3.91 (d, 1H, $J = 5.7$ Hz), 3.79 (s, 1H), 3.74 (t, 2H, $J = 5.4$ Hz), 2.58 (s, 1H), 1.92–2.00 (m, 1H), 1.88–1.89 (m, 2H), 1.78–1.86 (m, 1H), 1.44 (s, 9H), 0.91 (s, 9H) 0.07 (s, 3H), 0.06 (s, 3H). ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 170.9$, 169.0, 156.0, 136.6, 128.9, 128.5, 128.2, 82.1, 73.0, 72.6, 67.3, 61.2, 60.5, 59.6, 50.5, 47.3, 36.2, 29.4, 28.4, 26.3, 18.6, −5.07. HRMS (FAB) calcd. for $C_{29}H_{47}N_2O_8Si$ (MH^+) 579.3102, found 579.3123.

General procedure for the synthesis of ring closed compounds **10** *and 3,5-disubstituted proline derivatives* **11** *(GP 2)*

Dipeptides **8** (1.0 equiv.) were dissolved in a 2.5:1 mixture of acetone/water (15 mL per 5 mmol) and cooled to 0°C. $NaIO_4$ (2.0 equiv.) was added and the resulting mixture was stirred at 0°C for 2 h and then allowed to reach rt After stirring at rt for 20 min, brine was added (15 mL per 5 mmol of **8**) and the resulting aqueous solution was extracted with ethyl acetate. The organic phase was dried with Na_2SO_4, filtered, and the solvent was removed *in vacuo* to give colorless aldehydes **9** that were used without further purification. These aldehydes (1.0 equiv.) were dissolved in dry THF (30 mL per mmol aldehyde) and *tert*-butyl (triphenylphosphanylidene)

acetate (2.0 equiv.) was added. The resulting mixture was stirred at rt for 12 h. The solvent was removed *in vacuo* and the resulting crude product was purified by column chromatography (EA/PE, gradient) to give ring closed compounds **10** as well as proline derivatives **11** (see below for ratios) as colorless sticky solids.

Ring closed compound **10a** *and proline derivative* **11a**

The title compounds were synthesized according to GP 2 from 1.0 g (2.0 mmol) dipeptide **8a** to give **10a** in 590 mg (1.00 mmol, 50%) yield as well as **11a** in 490 mg (0.71 mmol, 36%) yield. **10a**: $R_f = 0.54$ (EA/PE, 1:1; Mo/Ce). $[\alpha]^{20}_D = +60.0$ (c = 0.21, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 6:4 mixture of rotamers): $\delta = 7.31$–7.38 (m, 5H), 6.81 (dd, 1H, $J = 7.6$, 15.4 Hz), 5.89–5.93 (m, 0.4H), 5.87 (d, 1H, $J = 15.5$ Hz), 5.70–5.73 (m, 0.6H), 5.11–5.24 (m, 2H), 4.70 (br, 0.4H), 4.64 (br, 0.6H), 4.13 (d, 1H, $J = 8.8$ Hz), 3.92–4.02 (m, 1H), 3.67 (s, 1.8H), 3.60 (s, 1.2H), 3.23 (dd, 0.6H, $J = 4.3$, 18.1 Hz), 3.20 (dd, 0.6H, $J = 2.6$, 18.1 Hz), 3.14 (dd, 0.4H, $J = 2.5$, 17.6 Hz), 2.98 (dd, 0.4H, 10-H, $J = 2.8$, 17.6 Hz), 2.88–2.95 (m, 1H), 2.12–2.19 (m, 2H), 1.48 (s, 9H), 1.45 (s, 9H). ^{13}C NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 165.1$, 164.6, 143.8, 128.8, 128.8, 128.7, 128.6, 128.3, 128.3, 125.6, 82.5, 81.9, 73.7, 73.3, 68.4, 68.3, 64.1, 62.01, 52.6, 52.5, 52.1, 51.9, 44.2, 37.2, 35.0, 34.0, 28.2, 28.1. HRMS (FAB) calcd. for $C_{30}H_{41}N_2O_{10}$ (MH^+) 589.2761, found 589.2752. **11a**: $R_f = 0.76$ (EA/PE, 1:1; Mo/Ce). $[\alpha]^{20}_D = +5.4$ (c = 1.0, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 7:3 mixture of rotamers): $\delta = 7.17$–7.26 (m, 5H), 6.71 (dd, 0.3H, $J = 7.4$, 15.5 Hz), 6.65 (dd, 0.7H, $J = 7.3$, 15.7 Hz), 6.63 (dd, 0.7H, $J = 7.4$, 15.8 Hz), 6.52 (dd, 0.3H, $J =$ 6.1, 15.5 Hz), 5.85 (dd, 0.7H, $J = 1.0$, 15.7 Hz), 5.81 (dd, 0.3H, $J = 1.0$, 15.5 Hz), 5.71 (dd, 0.7H, $J = 1.3$, 15.8 Hz), 5.57 (dd, 0.3H, $J = 1.3$, 15.7 Hz), 5.42 (d, 0.3H, $J = 8.1$ Hz), 5.36 (d, 0.7H, $J = 8.4$ Hz), 4.93–5.02 (m, 1.3H), 4.73–4.88 (m, 2.1H), 4.62–4.70 (m, 0.6H), 4.51 (dd, 0.3H, $J = 6.6$, 15.0 Hz), 4.19 (d, 0.7H, $J = 5.3$ Hz), 3.61 (s, 0.9H), 3.57 (s, 2.1H), 3.02–3.08 (m, 0.3H), 2.81–2.89 (m, 0.7H), 2.71 (d, 0.7H, $J = 6.5$ Hz), 2.69 (dd, 0.7H, $J = 6.2$, 15.8 Hz), 2.43–2.52 (m, 1.3H), 2.25–2.33 (m, 0.3H), 1.76–1.85 (m, 0.7), 1.88–1.76 (m, 0.3H), 1.30–1.38 (m, 27H). ^{13}C NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 171.1$, 170.8, 170.7, 169.2, 165.3, 165.1, 165.0, 164.6, 155.1, 155.1, 145.9, 145.5, 145.0, 144.8, 136.4, 128.6, 128.2, 128.1, 128.1, 125.2, 124.9, 123.1, 83.7, 82.2, 80.9, 80.7, 80.5, 67.0, 65.8, 65.8, 59.1, 58.8, 52.2, 50.1, 49.5, 45.6, 43.4, 39.0, 38.5, 38.1, 35.2, 28.2, 28.0, 27.9. HRMS (FAB) calcd. for $C_{36}H_{51}N_2O_{11}$ (MH^+) 687.3493, found 687.3517.

Ring closed compound **10b** *and proline derivative* **11b**

The title compounds were synthesized according to GP 2 from 1.56 g (2.92 mmol) dipeptide **8b** to give **10b** in 1.02 g (1.62 mmol, 55%) yield (Maison et al., 2004) as well as **11b** in 280 mg (0.38 mmol, 13%) yield. **11b**: $R_f = 0.68$ (EA/PE, 1:2; Mo/Ce). $[\alpha]^{20}_D = +9.1$ (c = 1.62, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 6:4 mixture of rotamers): $\delta = 7.24$–7.36 (m, 5H), 6.82 (dd, 0.6H, $J = 7.3$, 15.8 Hz), 6.74–6.80 (m, 0.4H), 6.73 (dd, 0.6H, $J = 7.3$, 15.5 Hz), 6.61 (dd, 0.4H, $J = 6.3$, 15.8 Hz), 5.97 (d, 0.4H, $J = 15.5$ Hz), 5.95 (dd, 0.6H, $J = 1.0$, 15.8 Hz), 5.81 (dd, 0.6H, $J = 1.3$, 15.8 Hz), 5.69 (dd, 0.4H, $J = 1.0$, 15.8 Hz), 5.38–5.45 (m, 1H), 5.00–5.14 (m, 1.6H), 4.81–4.98 (m, 2H), 4.73–4.79 (m, 0.4H), 4.51–4.57 (m, 0.4H), 4.30 (d, 0.6H, $J = 5.4$ Hz), 3.11–3.17 (m, 0.4H), 2.90–2.98 (m, 0.6H), 2.66–2.79 (m, 1.4H), 2.57 (dt, 0.6H, $J = 7.7$, 12.9 Hz), 2.35–2.44 (m, 1H), 1.88–1.95 (m, 0.6H), 1.81 (dt, 0.4H, $J = 2.8$, 12.9 Hz), 1.39–1.47 (m, 36H). ^{13}C NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 171.6$, 171.3, 170.4, 169.9, 169.3, 169.2, 165.4, 165.3, 165.1, 164.8, 155.2, 155.2, 145.9, 145.7, 145.1, 145.0, 136.7, 136.6, 128.7, 128.6, 128.2, 128.2, 128.1, 125.4, 125.2, 124.8, 123.3, 83.6, 82.3, 81.6, 81.0, 80.9, 80.7, 80.4, 80.4, 66.9, 65.8, 65.5, 59.1, 58.9, 50.1, 48.9, 45.7, 43.5, 40.4, 40.0, 38.2, 35.2, 28.4, 28.3, 28.2, 28.3, 28.2, 28.2, 28.0. HRMS (FAB) calcd. for $C_{39}H_{57}N_2O_{11}$ (MH^+) 729.3962, found 729.3985.

Ring closed compound **10c** *and proline derivative* **11c**

The title compounds were synthesized according to GP 2 from 2.00 g (3.65 mmol) dipeptide **8c** to give **10c** in 0.61 g (0.95 mmol, 26%) yield (Maison et al., 2004) as well as **11c** in 1.48 g (1.99 mmol, 55%) yield. **11c**: $R_f = 0.69$ (EA/PE, 1:2; Mo/Ce). $[\alpha]^{20}_D = -3.0$ (c = 1.0, $CHCl_3$). ^{1}H NMR (500 MHz, [D_4] MeOH, 8:2 mixture of rotamers): $\delta = 7.26$–7.37 (m, 5H), 6.74–6.90 (m, 1.8H), 6.77 (dd, 0.2H, $J = 6.3$, 15.5 Hz), 5.87–6.03 (m, 1H), 5.88 (dd, 0.8H, $J = 1.3$, 15.4 Hz), 5.74 (dd, 0.2H, $J = 1.3$, 15.8 Hz), 4.98–5.12 (m, 2.8H), 4.69–4.75 (m, 0.2H), 4.35–4.51 (m, 1H), 4.16–4.34 (m, 1H), 3.00–3.09 (m, 1H), 2.62 (dt, 0.8H, $J = 7.6$, 12.9 Hz), 2.31–2.46 (m, 2.2H), 2.04–2.13 (m, 1H), 1.85–1.96 (m, 0.6H), 1.62–1.73 (m, 1H), 1.35–1.54 (m, 36.4H). ^{13}C NMR (100 MHz, [D_4] MeOH, mixture of rotamers): $\delta = 174.1$, 174.0, 171.0, 166.7, 166.6, 165.7, 147.5, 147.2, 138.3, 129.4, 128.9, 128.9, 125.9, 125.8, 125.4, 83.3, 81.9, 81.9, 81.8, 67.5, 67.3, 60.1, 53.2, 44.7, 39.1, 31.5, 28.6, 28.5, 28.4, 28.4, 28.2, 28.2. HRMS (FAB) calcd. for $C_{40}H_{59}N_2O_{11}$ (MH^+) 743.4119, found 743.4157.

Ring closed compound **10d** *and proline derivative* **11d**

The title compounds were synthesized according to GP 2 from 2.50 g (4.33 mmol) dipeptide **8d** to give **10d** in 0.39 g (0.58 mmol, 13%) yield as well as **11d** in 1.83 g (2.37 mmol, 55%) yield. **10d**: $R_f = 0.56$ (EA/PE, 1:2; Mo/Ce). $[\alpha]^{20}_D = +30.1$ (c = 0.57, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 1:1 mixture of rotamers): $\delta = 7.30$–7.40 (m, 5H), 6.77–6.84 (m, 1H), 5.97 (t, 0.5H, $J = 2.9$ Hz), 5.87 (d, 0.5H, $J = 15.5$ Hz), 5.86 (d, 0.5H, $J = 15.5$ Hz), 5.83 (t, 0.5H, $J = 3.2$ Hz), 5.63 (d, 0.5H, $J = 3.5$ Hz), 5.34 (d, 0.5H, $J = 3.2$ Hz) 5.07–5.26 (m, 2H), 4.70 (t, 0.5H, $J = 4.7$ Hz), 4.64 (t, 0.5H, $J = 5.0$ Hz), 4.08–4.15 (m, 1H), 3.92–3.96 (m, 0.5H), 3.87–3.91 (m, 0.5H), 3.72–3.84 (m, 2H), 2.83–2.92 (m, 1H), 2.33–2.47 (m, 2H), 2.01–2.19 (m, 2H), 1.46 (s, 9H), 1.48 (s, 9H), 0.86 (s, 9H), 0.06 (s, 3H), 0.03 (s, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 172.9$, 166.4, 166.2, 144.5, 129.1, 129.08, 129.05, 128.9, 128.8, 128.7, 128.6, 125.7, 82.64, 82.59, 81.09, 81.07, 73.6, 73.3, 68.4, 68.3, 64.5, 62.5, 61.2, 61.1, 54.3, 54.0, 44.5, 34.1, 33.8, 32.3, 28.5, 28.4, 26.2, −5.1. HRMS (FAB) calcd. for $C_{35}H_{55}N_2O_9Si$ $(M + H)^+$ 675.3677, found 675.3669. **11d**: $R_f = 0.74$ (EA/PE, 1:2; Mo/Ce). $[\alpha]^{20}_D = +17.5$ (c = 0.44, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 7:3 mixture of rotamers): $\delta = 7.27$–7.39 (m, 5H), 6.62–6.87 (m, 2H), 5.74–5.94 (m, 2H), 4.92–5.27 (m, 2H), 4.67–4.83 (m, 1H), 4.57–4.67 (m, 0.7H), 4.44–4.53 (m, 0.3H), 4.31–4.34 (m, 0.7H), 4.11–4.15 (m, 0.3H), 3.62–3.83 (m, 2H), 3.14–3.19 (m, 0.3H), 2.85–2.97 (m, 0.7H), 2.54–2.61 (m, 0.7H), 2.35–2.44 (m, 0.3H), 2.03–2.18 (m, 0.3H), 1.88–2.00 (m, 1H), 1.76–1.87 (m, 0.7H), 1.67–1.75 (m, 0.7H), 1.54–1.63 (m, 0.3H), 1.42–1.50 (m, 27H), 0.85–0.92 (m, 9H), 0.02–0.11 (m, 6H). ^{13}C-NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 169.8$, 165.4, 165.2, 155.9, 145.4, 145.2, 136.8, 128.5, 128.1, 128.0, 125.1, 124.3, 123.5, 82.2, 80.9, 66.6, 65.7, 59.43, 59.4, 58.4, 50.2, 43.5, 37.9, 28.22, 28.19, 28.14, 28.09, 27.95, 26.2, 26.1, −5.0. HRMS (FAB) calcd. for $C_{41}H_{65}N_2O_{10}Si$ $(M + H)^+$ 773.4409, found 773.4450.

General procedure for the synthesis of ring closed compounds **12** *(GP 3)*

Proline derivatives **11** (1.0 equiv.) were dissolved in THF (20 mL per mmol **11**) and KO*t*Bu (4.0 equiv.) was added. After addition of water (2 mL per mmol **11**) the resulting solution was refluxed for 6 h and then stirred for 12 h at rt. The reaction was quenched with citric acid (10% in water; 20 mL per mmol **11**) and stirred for another hour at rt. A solution of NaOH (2M) was added to adjust to pH 10–12 before the mixture was extracted with ethyl acetate. The organic phase was dried with Na_2SO_4, filtered, and the solvent was removed *in vacuo*. This crude product was purified by column chromatography (EA/PE, gradient) to give **12** as colorless sticky solids.

Ring closed compound **12b**

The title compound was synthesized according to GP 3 from 110 mg (0.15 mmol) **11b** in 58.7 mg (0.08 mmol, 54%) yield. **12b**: $R_f = 0.37$ (EA/PE, 3:7; Mo/Ce). $[\alpha]^{20}_D = +20.8$ (c = 2.7, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 7.26$–7.39 (m, 5H), 6.74 (dd, 1H, $J = 7.3$, 15.4 Hz), 5.85 (d, 1H, $J = 15.4$ Hz), 5.12–5.23 (m, 3H), 5.04–5.10 (m, 1H), 4.11–4.17 (m, 1H), 4.07 (d, 1H, $J = 9.5$ Hz), 2.83–2.95 (m, 1H), 2.10–2.82 (m, 6H), 1.37–1.48 (m, 36H). ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 169.2$, 168.3, 165.8, 165.0, 156.3, 143.2, 137.1, 128.6, 128.3, 128.0, 125.7, 82.5, 81.8, 80.9, 80.8, 68.1, 63.9, 60.6, 52.9, 48.6, 44.1, 40.7, 35.0, 29.8, 28.2, 28.1, 28.1, 28.0. HRMS (FAB) calcd. for $C_{39}H_{57}N_2O_{11}$ $(M+H)^+$ 729.3962, found 729.3943.

Ring closed compound **12d**

The title compound was synthesized according to GP 3 from 0.41 mg (0.53 mmol) **11d** in 150 mg (0.19 mmol, 37%) yield. **12d**: $R_f = 0.31$ (EA/PE, 1:4; Mo/Ce). $[\alpha]^{20}_D = +16.2$ (c = 1.95, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 7:3 mixture of rotamers): $\delta = 7.27$–7.31 (m, 5H), 6.61–6.70 (m, 1H), 5.71–5.83 (m, 1H), 5.13–5.19 (m, 0.5H), 4.99–5.12 (m, 2.5H), 4.42–4.59 (m, 1H), 4.04–4.17 (m, 0.3H), 3.86–4.04 (m, 1.7H), 3.74–3.84 (m, 0.3H), 3.64–3.73 (m, 1H), 3.55–3.64 (m, 0.7H), 2.80–2.86 (m, 0.3H), 2.70–2.80 (m, 0.7H), 2.28–2.49 (m, 1.4H), 2.00–2.26 (m, 1.6H), 1.67–1.81 (m, 1H), 1.28–1.39 (m, 27H), 0.71–0.77 (m, 9H), −0.17 to −0.05 (m, 6H). ^{13}C-NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 169.4$, 166.7, 165.1, 154.89, 154.86, 144.0, 143.5, 136.4, 135.9, 128.6, 128.4, 128.2, 127.9, 127.8, 127.7, 127.1, 125.7, 125.5, 82.5, 82.3, 81.8, 81.7, 81.3, 80.9, 68.2, 67.5, 63.7, 63.6, 61.7, 61.3, 60.4, 60.3, 54.6, 53.0, 48.8, 48.1, 44.3, 44.0, 38.5, 37.9, 36.0, 35.1, 34.5, 34.3, 28.2, 28.1, 28.0, 26.0, −5.2, −5.3. HRMS (FAB) calcd. for $C_{41}H_{65}N_2O_{10}Si$ $(M+H)^+$ 773.4409, found 773.4412.

Ozonolysis of **12d** *to aldehyde* **13**

50.0 mg (64.7 μmol) **12d** were dissolved in 25 mL dry dichloromethane and cooled to −78°C. Ozone was bubbled through the solution for 5 min until a blue color persisted. The solution was purged with nitrogen for 2 min and then treated with dimethyl sulfide (0.84 g, 13.5 mmol) in 5 mL dry dichloromethane. The solution was allowed to reach rt and the solvent was evaporated *in vacuo*. The crude product was purified by column chromatography (EA/PE, gradient) to give the desired aldehyde containing minor impurities in 9.8 mg (14.5 μmol, 22%) yield as well as the TBDMS-deprotected compound **13** in another 10.0 mg (17.8 μmol, 28%) yield. **13**: $R_f = 0.20$ (EA/PE, 2:1; Mo/Ce). $[\alpha]^{20}_D = +7.2$ (c = 0.5, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 1:1 mixture of rotamers): $\delta = 9.74$ (s, 1H), 7.29–7.43 (m, 5H), 5.07–5.29 (m, 3H), 4.76 (dd, 0.5H, $J = 6.5$, 7.5 Hz), 4.68 (d, 1H, $J = 7.9$ Hz), 4.61 (dd, 0.5H, $J = 3.2$, 8.5 Hz), 4.08–4.20 (m, 1H), 3.59–3.74 (m, 2H), 2.99–3.07 (m, 1H), 2.45–2.56 (m, 1H), 2.35–2.44 (m, 0.5H), 2.14–2.33 (m, 2.5H), 1.68–1.83 (m, 2H), 1.47 (s, 9H), 1.41 (s, 9H). ^{13}C NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 197.1$, 196.9, 169.0, 168.7, 167.4, 155.6, 155.1, 134.8, 128.8, 128.8, 128.5, 128.5, 128.0, 128.2, 83.5, 82.1, 68.6, 61.3, 60.2, 59.7, 59.3, 55.5, 54.5, 52.3, 52.3, 49.1, 48.1, 38.8, 37.2, 34.4, 34.2, 29.2, 28.9, 28.1, 28.0. HRMS (FAB) calcd. for $C_{29}H_{41}N_2O_9$ $(M+H)^+$ 561.2812, found 561.2780.

Diazabicycloalkane **19d**

392 mg (0.58 mmol) of compound **10d** were dissolved in 12 mL dry MeOH and 25 mg of 5% Pd on activated charcoal were added. The suspension was stirred under an atmosphere of hydrogen for 18 h and subsequently filtered through a plug of celite. The solvent was removed under reduced pressure to give the appropriate secondary amine as pale brownish oil in 304 mg (0.58 mmol, 100%) yield: $R_f = 0.79$ (dichloromethane/MeOH, 19:1; Mo/Ce). $[\alpha]^{20}_D = -10.7$ (c = 1.0, $CHCl_3$). Without further purification, 283 mg (0.54 mmol) of this secondary amine were dissolved in 9 mL dry DMF and 135 mg (0.58 mmol) 6-(*tert*-butoxycarbonylamino)hexanoic acid as well as 232 mg (0.61 mmol) HATU were added. The resulting solution was cooled to 0°C and DIPEA (185 μL, 140 mg, 0.11 mmol) was added. The reaction mixture was stirred for 2 h, allowed to reach rt and stirred another 2.5 h at this temperature before the reaction was quenched by the addition of 200 mL water. The aqueous phase was extracted with dichloromethane (five times 70 mL) and the combined organic phases were washed with 2M HCl (100 mL). After separation, the organic layer was collected and the solvent was removed *in vacuo*. The residue was redissolved in diethylether and this solution was then washed once with 2M HCl (100 mL) and twice with a saturated solution of $NaHCO_3$ (each time 100 mL). The organic layer was dried ($NaSO_4$) and concentrated *in vacuo* to give the crude product that was purified by column chromatography (dichloromethane/MeOH, gradient) to give TBDMS-deprotected compound **19d** as colorless oil in 233 mg (0.37 mmol, 69%) yield. **19d**: $R_f = 0.32$ (dichloromethane/MeOH 95:5; Mo/Ce). $[\alpha]^{20}_D = +18.4$ (c = 0.5, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 7:3 mixture of rotamers): $\delta = 5.06$ (dd, 0.7H, $J = 4.4$, 9.8 Hz), 4.92 (dd, 0.3H, $J = 3.5$, 13.3 Hz), 4.70 (br, 0.3H), 4.63 (br, 0.7H), 4.56 (t, 0.3H, $J = 6.6$ Hz), 4.06 (dd, 0.7H, $J = 2.8$, 13.9 Hz), 4.02 (d, 0.7H, $J = 7.9$ Hz), 3.99 (d, 0.3H, $J = 7.6$ Hz), 3.80–3.87 (m, 1H), 3.69–3.79 (m, 0.6H), 3.61–3.65 (m, 0.7H), 3.43–3.50 (m, 0.7H), 3.06–3.11 (m, 2H), 2.95 (dd, 0.7H, $J = 11.5$, 13.9 Hz), 2.50 (dd, 0.3H, $J = 11.1$, 13.3 Hz), 2.14–2.43 (m, 7H), 1.94–2.06 (m, 1H), 1.85–1.93 (m, 0.3H), 1.70–1.78 (m, 1H), 1.61–1.70 (m, 2.7H), 1.42–1.52 (m, 2H), 1.46 (s, 9H), 1.43 (s, 9H), 1.42 (s, 9H), 1.30–1.37 (m, 2H), 1.17–1.24 (m, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 172.12$, 172.09, 172.06, 172.0, 170.5, 170.3, 167.5, 167.4, 156.2, 82.4, 82.3, 80.8, 80.7, 79.2, 64.6, 64.5, 58.9, 58.6, 58.3, 58.1, 55.2, 52.2, 45.7, 41.7, 41.6, 41.1, 40.4, 40.3, 36.5, 36.4, 36.0, 34.1, 33.7, 33.3, 29.9, 29.8, 29.3, 29.2, 28.5, 28.2, 28.1, 26.5, 26.4, 24.75, 24.68. HRMS (FAB) calcd. for $C_{32}H_{56}N_3O_9$ $(M+H)^+$ 626.4017, found 626.4011.

Diazabicycloalkane **19e**

240 mg (0.39 mmol) of the appropriate TBDMS-protected diazabicycloalkane (*tert*-butyl-((3*S*,6*S*,7*S*,9*S*)-2-benzoyl-7-(2-*tert*-butoxycarbonylethyl)-3-(*tert*-butyldimethyl-silyloxymethyl)-4-oxooctahydropyrrolo[1,2-*a*]pyrazin)-6-carboxylate) (Maison et al., 2004) were dissolved in 4 ml MeOH and 190 μL (188 mg, 1.17 mmol) triethylamin-trishydrofluorid and 0.33 ml (241 mg, 2.38 mmol) triethylamin were added. The solution was stirred at rt for 18 h, then heated to 40°C for 2.5 h and finally refluxed for 5 min. The solution was diluted with 3 mL MeOH and then washed with 5 mL of a saturated solution of $NaHCO_3$. The aqueous phase was extracted with dichloromethane and the combined organic phases were dried over Na_2SO_4. The solvent was removed *in vacuo*. The residue was redissolved in dichloromethane and the resulting solution was washed with a saturated solution of $KHSO_4$. The organic phase was dried (Na_2SO_4) and the solvent removed *in vacuo* to give **19e** as a colorless sticky solid containing minor impurities in 89.0 mg (0.18 mmol, 46%) yield. $R_f = 0.07$ (EA/PE 1:1; Mo-Ce). ^{1}H NMR (500 MHz, $CDCl_3$, 7:3 mixture of rotamers): $\delta = 7.47$–7.37 (m, 5H), 5.16 (br, 0.7H), 4.99 (d, 0.3H, $J = 12.0$ Hz), 4.38 (br, 0.3H), 4.25 (dd, 0.7H, $J = 3.8$, 11.0 Hz), 4.10–4.08 (m, 0.7H), 4.07 (d, 1H, $J = 7.6$ Hz), 4.03–3.97 (m, 0.7H), 3.96–3.80 (m, 1H), 3.29 (t, 0.7H, $J = 12.0$ Hz), 5.91 (t, 0.3H, $J = 11.2$ Hz), 2.37–2.25 (m, 1H), 2.24–2.16 (m, 1H), 2.13–2.06 (m, 0.6H), 2.05–1.96 (m, 1H), 1.78–1.65 (m, 1H), 1.50 (s, 9H), 1.42 (s, 9H), 1.34–1.25 (m, 1H), 1.13 (dd, 1.4H, $J = 11.4$, 23.4 Hz). ^{13}C NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 172.1$, 170.4, 166.0, 165.9, 135.3, 130.3, 129.7, 128.9, 128.5, 127.0, 82.5, 80.8, 64.6, 64.5, 63.8, 60.0, 58.5, 58.1, 56.7, 49.7, 43.4, 41.5, 36.2, 33.7, 29.0, 28.2, 28.1.

Target compound **20d**

83.9 mg (0.32 mmol) Ph_3P were dissolved in 1.5 mL dry THF, cooled to 0°C and stirred vigorously under an atmosphere of nitrogen while 62.2 μL (0.32 mmol) DIAD were added dropwise. The resulting suspension was stirred for 1 h before a solution of 100 mg (0.16 mmol) **19d** and 24.9 μL (0.35 mmol) AcSH in 1.5 mL dry THF was added. The solution was stirred at this temperature for 2 h, warmed to reach rt and stirred for 15 h at rt before the solvent was removed *in vacuo*. The resulting crude product was purified by column chromatography with EA/PE (gradient) to give **20d** in 70.4 mg (0.10 mmol, 64%) yield. **20d**: $R_f = 0.40$ (EA/PE 2:1; Mo/Ce). $[\alpha]^{20}_D = +5.6$ (c = 1.0, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 1:1 mixture of rotamers): $\delta = 5.15$ (dd, 0.5H, $J = 4.1$, 9.5 Hz), 4.98 (dd, 0.5H, $J = 3.5$, 13.2 Hz), 4.71 (br, 0.5H), 4.61 (br, 0.5H), 4.39 (t, 0.5H, $J = 6.9$ Hz), 4.06–4.11 (m, 0.5H), 4.00 (d, 0.5H, $J = 7.6$ Hz), 3.98 (d, 0.5H, $J = 7.9$ Hz), 3.82 (tt, 0.5H, $J = 4.7$, 11.0 Hz), 3.75 (tt, 0.5H, $J = 4.4$, 11.4 Hz), 2.92–3.14 (m, 4H), 2.80 (ddd, 0.5H, $J = 2.5$, 6.3, 9.5 Hz), 2.51 (dd, 0.5H, $J = 10.7$, 12.9 Hz), 2.15–2.46 (m, 10H), 1.86–2.05 (m, 2H), 1.64–1.76 (m, 3H), 1.48–1.55 (m, 2H), 1.47 (s, 4.5H), 1.46 (s, 4.5H), 1.44 (s, 9H), 1.43 (s, 9H), 1.32–1.40 (m, 2H), 1.14–1.22 (m, 1H). ^{13}C NMR (125 MHz, $CDCl_3$, mixture of rotamers): $\delta = 196.2$, 195.5, 172.1, 171.9, 170.6, 166.7, 82.4, 82.3, 80.8, 64.5, 64.4, 58.5, 57.9, 56.6, 53.8, 45.7, 41.7, 41.6, 41.3, 40.5, 40.5, 36.6, 36.5, 33.7, 33.4, 33.3, 32.9, 32.2, 30.8, 30.7, 30.0, 29.4, 29.3, 28.6, 28.2, 28.1, 28.1, 26.6, 26.2, 26.1, 25.0, 24.7. HRMS (FAB) calcd. for $C_{34}H_{58}N_3O_9S^+$ $(M+H)^+$ 684.3894, found 684.3874.

Target compound **20e**

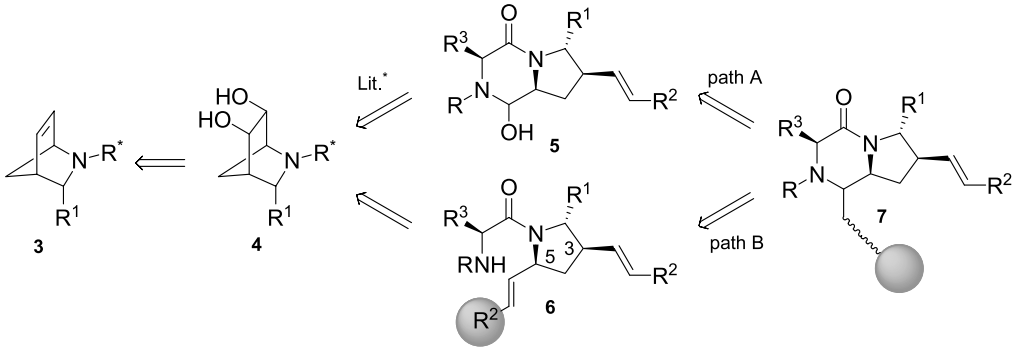
30.0 mg (0.11 mmol) Ph_3P were dissolved in 1 mL dry THF, cooled to 0°C and stirred vigorously under an atmosphere of nitrogen while 22.0 μL (0.11 mmol) DIAD were added dropwise. The resulting suspension was stirred for 30 min before a solution of 50.0 mg (0.10 mmol) **19e** and 8.50 μL (0.12 mmol) AcSH in 1 mL dry THF was added. The solution was stirred at this temperature for 2 h, warmed to reach rt and stirred for 1 h at rt before the solvent was removed *in vacuo*. The resulting crude product was purified by column chromatography (EA/PE, gradient) to give **20e** in 34.0 mg (0.06 mmol, 61%) yield. **20e**: $R_f = 0.17$ (EA/PE 1:2; Mo/Ce). $[\alpha]^{20}_D = +20.4$ (c = 1.35, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$, 7:3 mixture of rotamers): $\delta = 7.34$–7.46 (m, 5H), 5.43 (dd, 0.7H, $J = 3.5$, 6.9 Hz), 4.91–4.98 (m, 0.3H), 4.56 (br, 0.3H), 4.04 (d, 1H, $J = 8.5$ Hz), 3.95 (dd, 0.7H, $J = 3.2$, 13.2 Hz), 3.90–3.94 (m, 0.3H), 3.68–3.77 (m, 1.4H), 3.59 (dd, 0.7H, $J = 7.3$, 14.2 Hz), 3.25–3.43 (m, 0.6H), 3.19 (dd, 0.7H, $J = 10.7$, 13.2 Hz), 2.91 (t, 0.3H, $J = 12.0$ Hz), 2.24–2.37 (m, 5H), 2.13–2.23 (m, 1H), 2.03–2.10 (m, 1H), 1.95–2.02 (m, 1H), 1.66–1.81 (m, 1H), 1.49 (s, 2.7H), 1.48 (s, 6.3H), 1.44 (s, 2.7H), 1.41 (s, 6.3H), 1.08–1.17 (m, 1H). ^{13}C NMR (100 MHz, $CDCl_3$, mixture of rotamers): $\delta = 195.6$, 172.0, 170.4, 170.3, 165.3, 135.2, 130.2, 128.8, 127.1, 82.4, 80.7, 64.6, 58.5, 58.3, 58.2, 53.8, 48.8, 42.2, 41.5, 36.3, 33.6, 31.4, 30.7, 30.4, 29.0, 28.2, 28.1. HRMS (FAB) calcd. for $C_{29}H_{41}N_2O_7S$ $(M+H)^+$ 561.2634, found 561.2600.

Results

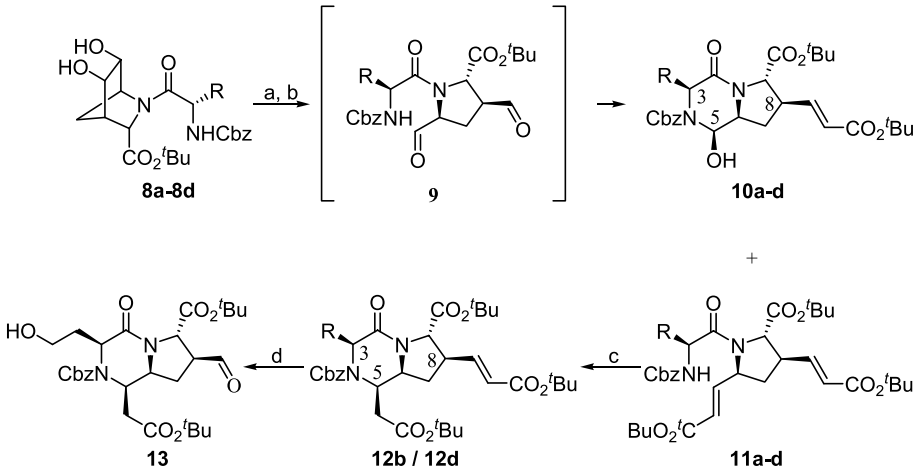
Besides the large number of studies directed to the synthesis of azabicycloalkanes of general type **2**, less effort has been put in the development of diazabicycloalkanes **1** (Fig. 1), presumably due to the high demands on a suitable synthetic route to these molecules. A suitable route has to be short and efficient with respect to the overall yield, it should be compatible with various side chain functionalities and should be stereoselective with respect to five stereocentres. Furthermore, a suitable synthesis should allow variations both in ring size as well as in stereochemistry. All of these demands were accomplished by a route that was recently published by our group. However, structural diversity of the target compounds **1** was limited with respect to the introduction of linker moieties at C-5 for conjugation to reporter groups and zinc complexing side chains at C-3 (Fig. 1). We needed both modifications of dipeptide mimetics **1** to obtain modular ligands for the cancer specific zinc protease PSMA.

Introduction of reporter groups

A retrosynthetic analysis of modular diazabicycloalkanes with a suitable linker moiety reveals two alternative routes (Scheme 1). Both routes are based on an oxidative cleavage of heterocycles like 4, a strategy that has been pioneered in peptide chemistry by Steglich and coworkers (Jaeger et al., 1995). A first approach would start with the conversion of the aminal function of an appropriate diazabicycloalkane intermediate **5** *via* *N*-acyl iminium chemistry (path A in Scheme 1). Similar conversions of cyclic amino acids *via* alkylation of *N*-acylated aminals are well known (see for example Maison et al., 2001;



Scheme 1. Retrosynthetic analysis of modular diazabicycloalkanes. *Maison et al., 2004



Scheme 2. Synthesis of modular diazabicycloalkane **13**. Residues R: **a**: $R = CH_2CO_2Me$; **b**: $R = CH_2CO_2{}^tBu$; **c**: $R = CH_2CH_2CO_2{}^tBu$; **d**: $R = CH_2CH_2COTBDMS$. *Reagents and conditions*: a) $NaIO_4$, acetone/H_2O, 0°C to rt, 30–60 min, b) $Ph_3P{=}CHCO_2C(CH_3)_3$, THF, rt, 12 h (13–55% over two steps for **10** as well as for **11**); c) KO^{t}Bu, THF/H_2O, reflux, 6 h, rt, 15 h (37–54%); d) O_3, DCM, −78°C (50%)

Wang et al., 2002; Collado et al., 1994; Plehiers et al., 2000; Polniaszek et al., 1990; Skrinjar et al., 1992; Ludwig and Wistrand, 1994; Speckamp and Moolenaar, 2000; Bloch, 1998). Aminal **5**, in turn, would be available *via* our previously described synthetic route with azabicyclodiol **4** as a key intermediate.

A second approach would start with 3,5-disubstituted proline derivative **6**, containing a linker moiety or an appropriate precursor R^2. Conversion to the final diazabicycloalkane scaffold **7** would be accomplished *via* an intramolecular Michael type addition (path B in Scheme 1). 3,5-disubstituted proline derivatives **6** would be synthesized from azabicyclodiol **4**, again *via* oxidative cleavage of **4** in the presence of a Wittig reagent.

In a first attempt we tried to attach suitable linker moieties via *N*-acyl iminium chemistry according to route A. Therefore, we tried to convert *N*-Boc protected aminals **5** (R = Boc) and derivatives thereof with various nucleophiles such as allyltrimethylsilane, copper organyls or cyanides in the presence of a number of different lewis acids with variations in reaction temperature and leaving group properties but no product could be isolated.

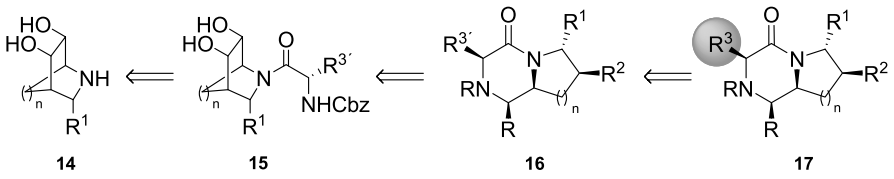
We were thus focusing on our second retrosynthetic alternative (path B in Scheme 1). 3,5-disubstituted proline derivatives **6** are key intermediates for this second route to modular diazabicycloalkanes with a suitable linker moiety. As we have shown before, bishydroxylated dipeptides **8** are valuable precursors for diazabicycloalkanes of general type **1**. Upon oxidative cleavage these diols give intermediate bisaldehydes **9** (Scheme 2) that cyclize to give the desired diazabicycloalkane scaffold with intramolecular aminal formation. These intermediate aminals (not shown) can be converted by a subsequent Wittig reaction to different dipeptide mimetics **10a**–**10d** (Maison et al., 2002, 2004). However, in some cases, if the steric demand of R is high, cyclisation of the intermediate **9** is slow and the bisaldehyde **9** can be trapped by a Wittig reagent to give 3,5-disubstituted proline derivatives like **11a**–**11d** (Scheme 2).

This unexpected finding turned out to be a valuable step on the way to modular ligands for PSMA and we were happy to learn that conversion of compounds **11** with potassium-*tert*-butylate leads to bicyclic diazabicycloalkanes **12b** and **12d** *via* ring closure in a diastereoselective Michael type reaction. The relative configuration of aminals **10a**–**d** and diazabicycloalkane **12b** and **12d** was determined by 2D-NOESY spectra. Strong NOE crosspeaks were observed for 3-H, 5-H and 8-H. The exclusive formation of 3,5-*cis* substituted diazabicycloalkanes **10a**–**d**, **12b** and **12d** is most likely a consequence of pseudoallylic 1,3-strain (Seebach et al., 1992), that would be apparent in the corresponding 3,5-*trans* derivatives.

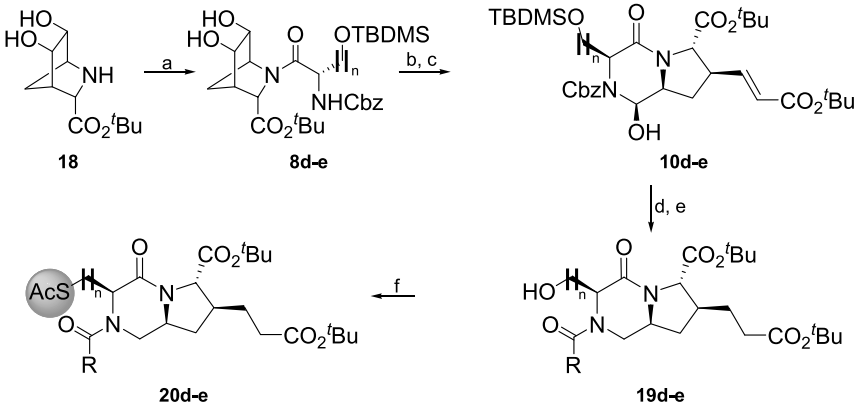
Olefin **12d** was converted into aldehyde **13** by ozonolysis. Aldehyde **13** is a versatile intermediate for a number of dipeptide mimetics, since the aldehyde function can be converted to various amino acid side chains according to our previously published protocols (Maison et al., 2002, 2004).

Introduction of zinc complexing side chains

As peptides with Zn^{2+} complexing, non-ionic side chains such as methionine or cysteine show high affinity for



Scheme 3. Retrosynthetic analysis of sulphur containing azabicycloalkanes **17**



Scheme 4. Synthesis of Cys-*H*Glu (n = 1, compounds "**e**") and *H*Cys*H*Glu (n = 2, compounds "**d**") mimetics **20**. Residues R: **d**: R = $(CH_2)_5NHBoc$; **e**: R = C_6H_5. *Reagents and conditions*: a) Cbz-*N*-protected amino acid (Ser, *H*Ser), DCC, HOBt, DMF, 12 h, 0°C – rt (53–59%); b) $NaIO_4$, acetone/H_2O, 0°C – rt, 30–60 min, c) $Ph_3P=CHCO_2C(CH_3)_3$, THF, rt, 12 h (13*–43% over two steps (*the low yield for compound **10d** was due to slow cyclisation. In consequence, bisolefin **11d** was formed as major product; for **11d** compare with Scheme 2)); d) H_2 (1atm), 5% Pd/C, MeOH, rt, 24 h (92–100%); e) for conversion of **d**: $BocNH(CH_2)_5CO_2H$, HATU, DIPEA, DMF (69%); f) DIAD, Ph_3P, AcSH, THF, 0°C – rt (61–64%); for conversion of **e**: 1. BzCl, DIPEA, DCM (77%); 2. $Et_3N \cdot (HF)_3$, MeOH (46%)

PSMA we were interested in introducing sulphur-containing amino acid side chains into the diazabicycloalkane scaffold.

Again, two general retrosynthetic analyses are possible. The most obvious synthetic route would start with a coupling reaction of an appropriate sulphur containing *N*-terminal amino acid to the azabicyclodiol **14** in order to give dipeptides that could be further converted to the diazabicycloalkane scaffold by oxidative cleavage of the diol and subsequent intramolecular aminal formation according to our standard protocol depicted in Scheme 2. However, the sulphur containing side chains appeared to be susceptible to oxidation in this synthetic step, too.

Alternatively, one could introduce sulphur into the side chain of the *N*-terminal amino acid (R^3) after complete formation of the diazabicycloalkane scaffold by conversion of appropriate precursors $R^{3'}$ in **16** to R^3 in **17**. A suitable precursor in side chain $R^{3'}$ could be a primary alcohol that would be transformed to a thiol group *via* Mitsunobu reaction. Intermediate **16**, in turn, could be derived from dipeptides **15** (Scheme 3).

Starting with the conversion of **18** and *N*-Cbz-protected L-Serine or L-Homoserine under standard coupling conditions, dipeptides **8d** and **8e** were obtained in moderate to good yields. Oxidative cleavage of these intermediates and subsequent Wittig reaction lead to diazabicycloalkanes **10d** and **10e**. Hydrogenolysis followed by acylation of the resulting secondary amine yielded diazabicycloalkanes **19d** and **19e**. The primary alcohol of these Ser-*H*Glu and *H*Ser-*H*Glu mimetics was converted *via* thio-Mitsunobu reaction to give thioacetates **20d** and **20e**, protected Cys-*H*Glu and *H*Cys-*H*Glu mimetics, in good yield (Scheme 4).

Discussion

In summary, we have presented a short synthetic route to modular and polyfunctional diazabicycloalkanes *via* a new ring closing methodology. Sterically hindered 3,5-disubstituted proline derivatives **11** are key intermediates in this synthesis providing access to diazabicycloalkanes **12** and **13** with linker moieties for coupling to other functional molecules such as reporter groups. In comparison with modular diazabicycloalkanes described previously, these new derivatives might be advantageous for applications in combinatorial chemistry. Furthermore, we have

reported the efficient incorporation of sulphur containing side chains in our dipeptide mimetics. Detailed studies on structure activity relationships with respect to PSMA binding are currently in progress.

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