

# A concise total synthesis of salinosporamide A

Nicholas P. Mulholland,<sup>a</sup> Gerald Pattenden<sup>\*a</sup> and Iain A. S. Walters<sup>b</sup>

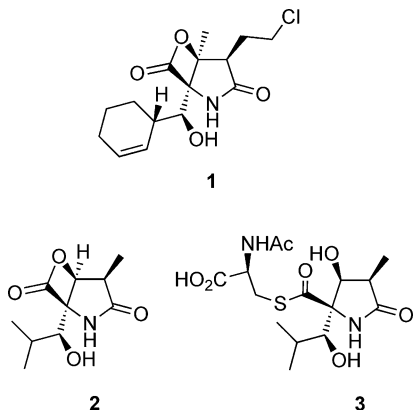
Received 19th May 2006, Accepted 12th June 2006

First published as an Advance Article on the web 29th June 2006

DOI: 10.1039/b607109k

A concise and straightforward 14-step total synthesis of (±)-salinosporamide A, based on a diastereoselective acid-catalysed intramolecular cyclisation of **6** to the pyrrolidinone **7**, and a regioselective reduction of the malonate derivative **8b** to the aldehyde **9**, is described.

Salinosporamide A (**1**) is a potent proteasome 20S inhibitor recently isolated from a marine bacterium of the new genus *Salinospora*, by Fenical *et al.*<sup>1</sup> The metabolite is unique, but is related to the β-lactone pyrrolidinone-based terrestrial natural product omuralide (also known as *clasto*-lactacystin β-lactone) **2**,<sup>2</sup> which is formed by lactonisation of the more familiar proteasome 20S inhibitor lactacystin **3**.<sup>3</sup> Salinosporamide A is reported to be approximately thirty five times more effective at proteasome inhibition than omuralide. The special proteasome inhibitory properties of the natural pyrrolidinones **1**, **2** and **3** have heightened interest in their potential in therapy for various types of cancer, also Alzheimer's disease, arthritis and asthma. It is not surprising therefore that these compounds have been attractive targets for total synthesis.<sup>4</sup>



The first total synthesis of salinosporamide A (**1**) was described by Corey *et al.*,<sup>5</sup> using a route starting from *S*-threonine. A year later Corey *et al.*<sup>6</sup> published a modified route to salinosporamide A from *S*-threonine and, simultaneously, Danishefsky *et al.*<sup>7</sup> presented an alternative synthesis of the natural product starting from a known chiral pool pyroglutamate derivative.<sup>8</sup> In earlier investigations we described a synthetic route to (+)-lactacystin **3**, which was based on a novel radical cyclisation of an α-ethynyl substituted serine as the key step.<sup>9</sup> We now present a concise and straightforward 14-step total synthesis of (±)-salinosporamide A, which is outlined in Scheme 1. Our synthesis of **1** hinges

on: i) a stereocontrolled acid catalysed intramolecular cyclisation of the substituted amide **6** leading to the pyrrolidinone **7**, ii) a regioselective reduction of the malonate derivative **8b** producing the key aldehyde intermediate **9**, and iii) installation of the cyclohexenyl side chain, from **9**. Although no biosynthesis studies have been published, our synthetic approach has features in common with the most likely origin of the pyrrolidinone ring in salinosporamide A *in vivo*, *i.e.* an intramolecular aldolisation from a substituted β-keto amide intermediate derived from a β-keto acid and an α-amino acid.<sup>10,11</sup>

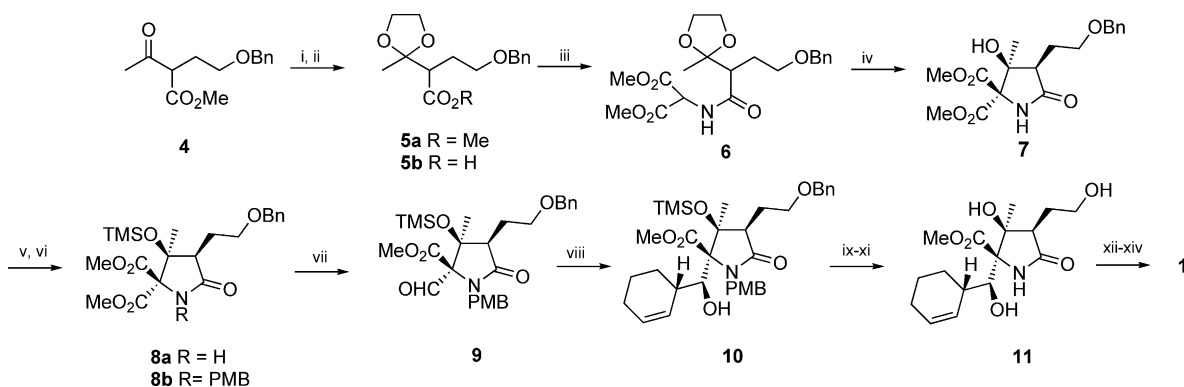
Thus, protection of the α-substituted β-keto ester **4**<sup>12</sup> as its dioxolan **5a**, followed by hydrolysis to the corresponding carboxylic acid **5b** and treatment with dimethyl 2-aminomalonate first gave the substituted amide **6**. When a solution of **6** in 4 : 1 acetic acid–water<sup>13</sup> was heated at 65 °C for 4 days, it underwent deprotection of the dioxolan and *in situ* intramolecular cyclisation leading to a single diastereomer of the (±)-pyrrolidinone **7**, which was obtained as colourless crystals, mp 82–83 °C. X-Ray crystallographic analysis showed that the pyrrolidinone had the expected *anti* arrangement between the C3–C4 alkyl chains shown in structure **7**.<sup>14</sup> Treatment of the tertiary alcohol **7** with excess TMSOTf in CH<sub>2</sub>Cl<sub>2</sub> containing 2,6-lutidine at –78 °C to 0 °C, followed by 1 M HCl, gave the corresponding TMS ether **8a** in 91% yield.<sup>15</sup> The nitrogen centre in the pyrrolidinone **8a** was next protected as its PMB derivative **8b** which, to our satisfaction, underwent regioselective reduction using Super-hydride in CH<sub>2</sub>Cl<sub>2</sub> at –78 °C producing the aldehyde **9** in 78% yield.<sup>16</sup>

The stage was now set to carry out the difficult operation of attaching the 2-cyclohexenyl side-chain to the aldehyde group in **9**, at the same time installing the correct stereochemistry for the newly introduced stereogenic centres. Fortunately, an elegant solution to this problem had already been worked out by Corey *et al.*,<sup>5</sup> and this protocol was also followed by Danishefsky *et al.*, in their synthesis of salinosporamide A. Thus, the aldehyde **9**, was treated with 2-cyclohexenylzinc bromide in THF at –78 °C, according to the protocol of Corey *et al.*, and we were delighted to find that the addition was essentially diastereoselective producing the adduct **10** in 87% yield.<sup>17</sup> Sequential deprotection of the TMS and benzyl ether groups in **10**, followed by the PMB group, then gave the triol ester **11**, which is the same intermediate in the synthesis of salinosporamide A presented by Corey *et al.*,<sup>5</sup> Hydrolysis of the methyl ester in **11**, followed by treatment of the resulting β-hydroxy acid, *in situ*, with BOP-Cl to give the corresponding β-lactone, and then chlorination with Ph<sub>3</sub>PCl<sub>2</sub> finally gave (±)-salinosporamide A (**1**), as a colourless solid, mp 169–172 °C. The synthetic salinosporamide A showed <sup>1</sup>H and <sup>13</sup>C NMR spectroscopic data and mass spectrometric data which were identical to those presented for the natural product.

We have therefore developed a conceptually straightforward synthetic route to (±)-salinosporamide A (**1**), which uses 14 steps

<sup>a</sup>School of Chemistry, University of Nottingham, Nottingham, NG7 2RD, England, UK. E-mail: GP@nottingham.ac.uk; Tel: +44 (0)115 951 3530

<sup>b</sup>AstraZeneca R & D Charnwood, Medicinal Chemistry, Bakewell Road, Loughborough, LE11 5RH, England, UK



**Scheme 1** Reagents and conditions: (i) ethylene glycol, *p*-TSA, PhH, 110 °C, 14 h; (ii) 2 M NaOH, EtOH, 70 °C, 3 h; (iii) dimethyl aminomalonate.HCl, HOBT, EDC.HCl, CH<sub>2</sub>Cl<sub>2</sub>, NMM, 0 °C to RT (82% over 3 steps); (iv) 4 : 1 AcOH–H<sub>2</sub>O, 65 °C, 4 days (71%); (v) excess TMSOTf, 2,6-lutidine, CH<sub>2</sub>Cl<sub>2</sub>, –78 °C to 0 °C, then 1 M HCl (91%); (vi) PMB–Br, NaH, DMF, 0 °C to RT, 14 h (82%); (vii) Super-hydride (1.0 M in THF), CH<sub>2</sub>Cl<sub>2</sub>, –78 °C, 3 h (78%); (viii) 2-cyclohexenylzinc bromide, THF, –78 °C (87%); (ix) BCl<sub>3</sub>.DMS, CH<sub>2</sub>Cl<sub>2</sub>, 24 h, 0 °C to RT; (x) 48% HF in H<sub>2</sub>O–MeCN (1 : 9), RT, 22 h; (xi) CAN, MeCN, H<sub>2</sub>O (3 : 1), 0 °C, 1 h (87% over 3 steps); (xii) [MeTeAlMe<sub>2</sub>], PhMe, RT, 24 h; (xiii) BOP–Cl, CH<sub>2</sub>Cl<sub>2</sub>, pyridine, RT, 3 h; (xiv) PPh<sub>3</sub>Cl<sub>2</sub>, MeCN, pyridine, RT, 4 h (45% over 3 steps).

from the substituted  $\beta$ -ketoester **4**. The key features of the synthetic route are the diastereoselective acid-catalysed cyclisation of **6** to **7**, and the facile regioselective reduction of the malonate **8b** to the aldehyde **9**, using Super-hydride at –78 °C. There are a range of options available to develop the route into an enantioselective synthesis of (+)-salinosporamide A, and some of those options are now being pursued.

We thank AstraZeneca for financial support (studentship to N. P. M.). We also thank Dr A. J. Blake of this School for the crystal structure determination of **7**.

## Notes and references

- R. H. Felting, G. O. Buchanan, T. J. Mincer, C. A. Kauffman, P. R. Jensen and W. Fenical, *Angew. Chem., Int. Ed.*, 2003, **42**, 355–357.
- Reviewed in: (a) E. J. Corey and W.-D. Z. Li, *Chem. Pharm. Bull.*, 1999, **47**, 1–10; (b) E. J. Corey, G. A. Reichard and R. Kania, *Tetrahedron Lett.*, 1993, **34**, 6977–6980; (c) E. J. Corey and G. A. Reichard, *J. Am. Chem. Soc.*, 1992, **114**, 10677–10678; (d) G. Fenteany, R. F. Standaert, G. A. Reichard, E. J. Corey and S. L. Schreiber, *Proc. Natl. Acad. Sci. U. S. A.*, 1994, **91**, 3358–3362.
- (a) S. Omura, T. Fujimoto, K. Otoguro, K. Matsuzaki, R. Moriguchi, H. Tanaka and Y. Sasaki, *J. Antibiot.*, 1991, **44**, 113–116; (b) S. Omura, K. Matsuzaki, T. Fujimoto, K. Kosuge, T. Furuya, S. Fujita and A. Nakagawa, *J. Antibiot.*, 1991, **44**, 117–118.
- For a review see: J. S. Panek, C. E. Masse, A. J. Morgan and J. Adams, *Eur. J. Org. Chem.*, 2000, 2513–2528, and references therein; see also (a) T. J. Donohoe, H. O. Sintim, L. Sisangia, K. W. Ace, P. M. Guyo, A. Cowley and J. D. Harling, *Chem.–Eur. J.*, 2005, **11**, 4227–4238; (b) T. J. Donohoe, H. O. Sintim, L. Sisangia and J. D. Harling, *Angew. Chem., Int. Ed.*, 2004, **43**, 2293–2269; (c) H. Ooi, N. Ishibashi, Y. Iwabuchi, J. Ishihara and S. Hatekeyama, *J. Org. Chem.*, 2004, **69**, 7765–7768; (d) J. J. Wardrop and E. G. Bowen, *Chem. Commun.*, 2005, 5106–5108; (e) C. J. Hayes, A. E. Sherlock and M. D. Selby, *Org. Biomol. Chem.*, 2006, **4**, 193–195; (f) N. Fukuda, K. Sasaki, T. V. R. S. Sastry, M. Kanai and M. Shibasaki, *J. Org. Chem.*, 2006, **71**, 1220–1225.
- L. R. Reddy, P. Saravanan and E. J. Corey, *J. Am. Chem. Soc.*, 2004, **126**, 6230–6231.
- (a) L. R. Reddy, J.-F. Fournier, B. V. S. Reddy and E. J. Corey, *Org. Lett.*, 2005, **7**, 2699–2701; (b) L. R. Reddy, J.-F. Fournier, B. V. S. Reddy and E. J. Corey, *J. Am. Chem. Soc.*, 2005, **127**, 8974–8976.
- A. Endo and S. J. Danishefsky, *J. Am. Chem. Soc.*, 2005, **127**, 8298–8299.
- cf.* (a) J. K. Thottathil, J. L. Moniot, R. H. Mueller, M. K. T. Wong and T. P. Kissick, *J. Org. Chem.*, 1986, **51**, 3140–3143; (b) Y. Hamada, A. Kawai, Y. Kohno, O. Hara and T. Shioiri, *J. Am. Chem. Soc.*, 1989, **111**, 1524–1525; (c) Y. Hamada, O. Hara, A. Kawai, Y. Kohno and T. Shioiri, *Tetrahedron*, 1991, **47**, 8635–8652.
- C. J. Brennan, G. Pattenden and G. Rescourio, *Tetrahedron Lett.*, 2003, **44**, 8757–8760.
- For some studies on the biosynthesis of lactacystin **3**, see: A. Nakagawa, S. Takahashi, K. Uchida, K. Matsuzaki, S. Omura, A. Nakamura, N. Kurihara, T. Nakamatsu, Y. Miyake, K. Take and M. Kainosho, *Tetrahedron Lett.*, 1994, **35**, 5009–5012.
- Some studies of the biosynthesis of salinosporamide A have been made by L. L. Beer and B. S. Moore; unpublished work, personal correspondence with B. S. Moore, Scripps Institution of Oceanography, UCSD, CA.
- cf.* M. Lee and D. H. Kim, *Bioorg. Med. Chem.*, 2002, **10**, 913–922.
- For a related acid catalysed reaction of diketene with substituted aminomalonates see: G. Simig, G. Doleschall, G. Hornyák, J. Fetter, K. Lempert, J. Nyitrai, P. Huszthy, T. Gizur and M. Kajtár-Peregy, *Tetrahedron*, 1985, **41**, 479–484.
- Crystal data for **7**: C<sub>18</sub>H<sub>23</sub>NO<sub>7</sub>, *M* = 365.37, monoclinic, *a* = 14.292(6), *b* = 11.057(4), *c* = 11.287(5) Å,  $\beta$  = 92.090(7)°, *U* = 1782.5(13) Å<sup>3</sup>, *T* = 150(2) K, space group *P2*<sub>1</sub>/*c*, *Z* = 4,  $\mu$ (Mo–*K* $\alpha$ ) = 0.105 mm<sup>–1</sup>, 15038 reflections measured, 4068 unique (*R*<sub>int</sub> = 0.124). Final *R*<sub>1</sub> [3308 *F* ≥ 4 $\sigma$ (*F*)] = 0.0421, *wR*<sub>2</sub> (all data) = 0.123. CCDC reference number 608046. For crystallographic data in CIF or other electronic format see DOI: 10.1039/b607109k.
- A small amount (<10%) of the corresponding *O*-TMS epimer was produced concurrently, resulting from a retro-aldolisation process. The epimer was cleanly removed by chromatography.
- The regioselective reduction of **8b**, leading to **9**, can be rationalised on steric grounds, with the bulky *O*-TMS group inhibiting hydride delivery to the adjacent *syn*-orientated CO<sub>2</sub>Me group. It is also possible that the same *O*-TMS group exercises an inductive effect and activates the corresponding *anti*-orientated CO<sub>2</sub>Me to reduction, as a consequence of their antiplanar relationship.
- Data for compound **10**: colourless solid, mp 157–160 °C;  $\nu_{\max}$  (CHCl<sub>3</sub>)/cm<sup>–1</sup> 3564, 2953, 1755, 1721, 1688, 1514;  $\delta_{\text{H}}$  (360 MHz, CDCl<sub>3</sub>) 7.34–7.26 (5H, m, C<sub>6</sub>H<sub>5</sub>), 7.23 (2H, d, *J* 8.7, *ArH*), 6.80 (2H, d, *J* 8.7, *ArH*), 6.05 (1H, app. d, *J* 10.2, CH<sub>2</sub>CH=), 5.63 (1H, app. d, *J* 10.2, CH<sub>2</sub>CH=CH), 4.80 (1H, d, *J* 15.3, OCHHPMB), 4.52 (2H, s, OCH<sub>2</sub>Ph), 4.42 (1H, d, *J* 15.3 OCHHPMB), 4.20 (1H, dd, *J* 3.3, 7.9, CH(OH)), 3.89–3.80 (2H, m, CH<sub>2</sub>OBn), 3.79 (3H, s, CO<sub>2</sub>Me), 3.62 (3H, s, ArOMe), 3.03 (1H, dd, *J* 3.8, 9.4, C(=O)CH), 2.26 (1H, br s, CH<sub>2</sub>CH=CHCH), 2.04 (2H, br s, CH<sub>2</sub>), 1.91–1.89 (2H, m, CH<sub>2</sub>), 1.76 (3H, s, CCH<sub>3</sub>), 1.59–1.51 (2H, m, CH<sub>2</sub>), 0.16 (9H, s, TMS);  $\delta_{\text{C}}$  (90 MHz, CDCl<sub>3</sub>) 177.7 (s), 169.5 (s), 157.8 (s), 138.7 (s), 134.7 (d), 130.6 (s), 128.2 (d), 127.7 (d), 127.3 (d), 127.1 (d), 123.8 (d), 113.2 (d), 86.2 (s), 82.4 (s), 76.8 (d), 72.9 (t), 68.8 (t), 55.2 (q), 51.7 (q), 48.3 (d), 47.7 (t), 38.2 (d), 29.4 (t), 26.1 (t), 25.0 (t), 20.8 (q), 20.5 (t), 2.7 (q); *m/z* (ES) Found 632.3041 (*M* + H<sup>+</sup>, C<sub>34</sub>H<sub>47</sub>NO<sub>7</sub>SiNa requires 632.3014).