Enantioselective Synthesis of Aurilide, a Cytotoxic 26-Membered Cyclodepsipeptide of Marine Origin

Tsuyoshi Mutou, Kiyotake Suenaga, Tatsuya Fujita, Takashi Itoh, Noboru Takada, Kozue Hayamizu, Hideo Kigoshi, and Kiyoyuki Yamada*

Department of Chemistry, Faculty of Science, Nagoya University, Chikusa, Nagoya 464, Japan

Fax +81 52 789 5041; e-mail yamada@chem3.chem.nagoya-u.ac.jp

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Abstract: Aurilide (**1**), a novel cyclodepsipeptide isolated from the Japanese sea hare *Dolabella auricularia*, was enantioselectively synthesized, and the present result unambiguously confirmed its stereostructure. In addition, the cytotoxicity of **1** was evaluated by employing synthetic **1**.

Recently, we isolated aurilide (**1**) as a very minute constituent from the Japanese sea hare *Dolabella auricularia* and elucidated the absolute stereostructure of **1** on the basis of its spectral analysis and organic synthetic methods.[1] Although aurilide (**1**) was isolated from a cytotoxic fraction of the sea hare, the scarcity of a natural supply has prevented the evaluation of its cytotoxicity. This fact and the novel 26-membered cyclodepsipeptide structure prompted us to synthesize aurilide (**1**). We describe herein the enantioselective synthesis of aurilide (**1**) and its biological evaluation.

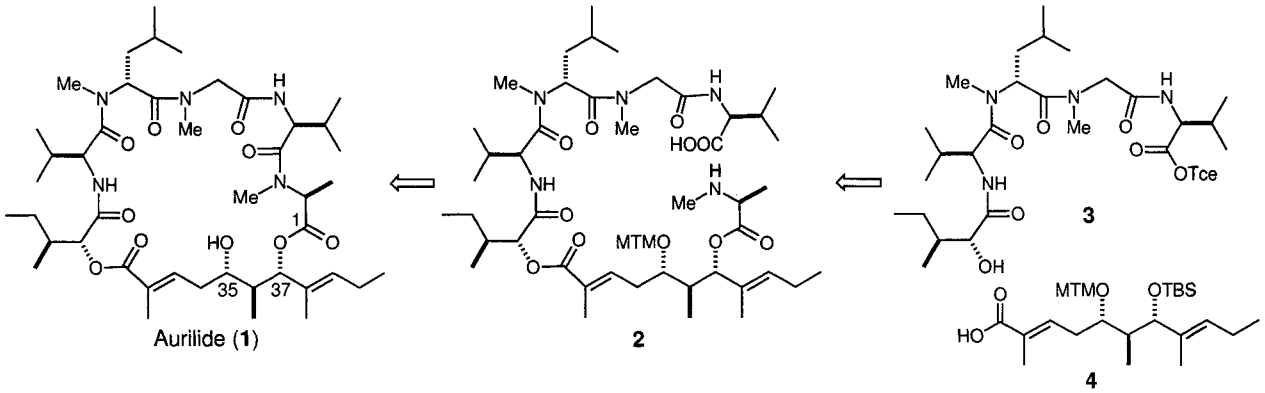
A key step in the synthesis of aurilide (**1**) is the 26-membered ring closure (Scheme 1). After careful inspection of the aurilide structure (**1**), we planned to construct the cyclic structure of **1** by the macrolactamization of amino acid **2**, which is synthesized from pentapeptide **3** and the protected dihydroxy acid **4**.

The synthesis of pentapeptide **3** was carried out starting from *N*-methylglycine *tert*-butyl ester hydrochloride in a stepwise manner in 84% overall yield (Scheme 2).

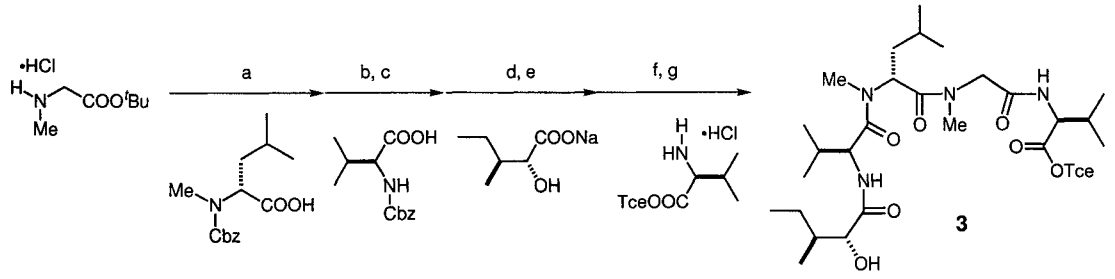
The protected dihydroxy acid **4** was synthesized from (4*S*,5*R*)-4-methyl-5-phenyl-3-propionyl-2-oxazolidinone (**5**), as shown in Scheme 3. The *anti* selective aldol reaction[2] between **5** and *trans*-2-methyl-2-pentenal afforded aldol **6**[3,4] (67%), which was converted into aldehyde **7** by the standard synthetic reactions (78%, 3 steps). The vinylogous Mukaiyama aldol reaction[5] between **7** and 2-methyl-1-trimethylsiloxy-1,3-butadiene provided a conjugated aldehyde, oxidation of which and subsequent treatment with diazomethane gave methyl ester **8** as a single diastereomer (59%, 3 steps).[6] Configuration inversion of the C35 hydroxyl group in **8** was effected as follows: Dess-Martin oxidation[7] of **8** afforded keto ester **9**, and reduction of the resulting keto group in **9** stereoselectively proceeded to give alcohol **10** (81% from **8**),[8] which has the desired stereochemistry concerning the C35 hydroxyl group.[6] Protection of the hydroxyl group in **10** followed by hydrolysis provided the protected dihydroxy acid **4** (68%, 2 steps).

The coupling reaction of pentapeptide **3** and the protected dihydroxy acid **4** was effected with EDCI•HCl[9] and DMAP to provide ester **11**, which was converted into alcohol **12** (91% from **4**). Esterification of **12** with *N*-Fmoc-*N*-methyl-L-alanine gave the *N*-methylalanine ester **13** (92%). The trichloroethyl group and the Fmoc group in **13** were removed in two steps to give amino acid **2**, which was subjected to macrolactamization with Bop-Cl[10] to afford cyclodepsipeptide **15** (29% from **13**).[11] Finally, the MTM group in **15** was removed to give aurilide (**1**) in 93% yield. Synthetic aurilide (**1**) was found to be identical with the natural product **1** by comparison of their spectral (UV, IR, ^{1}H NMR, MS, and α_D) and chromatographic properties.

In conclusion, the enantioselective synthesis of aurilide (**1**) was carried our in 3.9% overall yield based on the longest linear sequence (18 steps),[15] and the stereostructure of aurilide was unambiguously confirmed to be **1**. Aurilide (**1**) was found to exhibit a strong cytotoxicity against HeLa S_3 cells with an IC_{50} of 0.011 μg/mL by employing the synthetic specimen.

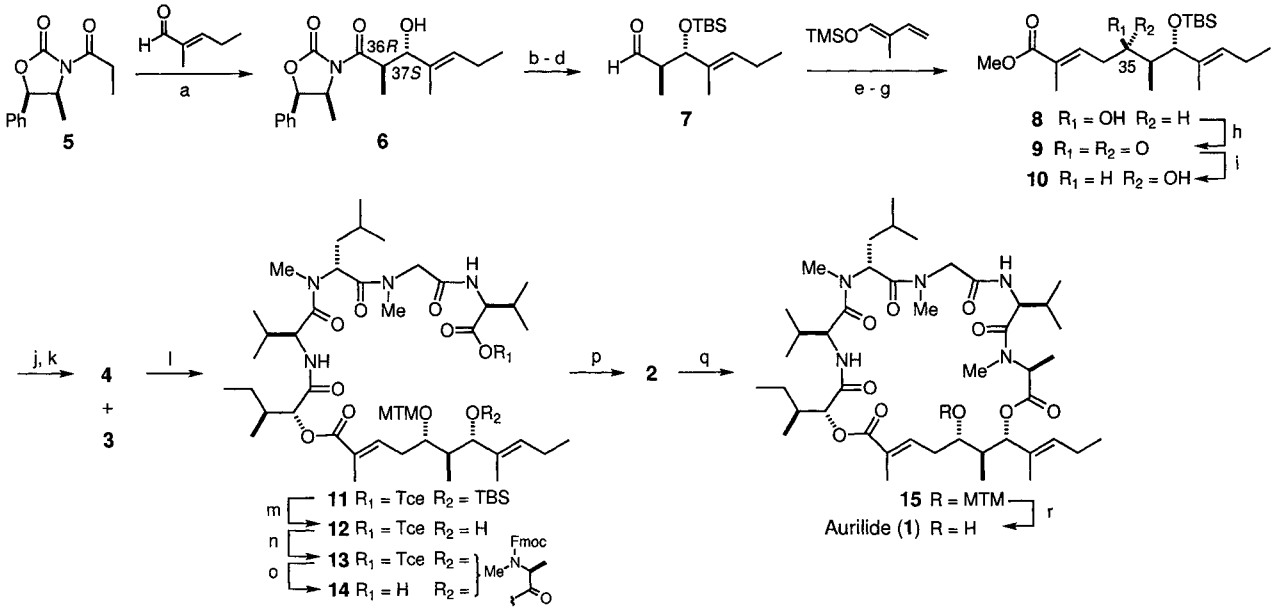


Scheme 1



Reagents and conditions: (a) DEPC,[13] Et_3N, DMF, 23 °C (98%); (b) H_2, Pd-C, EtOH, 23 °C; (c) PyBOP,[14] *i*-Pr_2NEt, CH_2Cl_2, 23 °C (92%, 2 steps); (d) H_2, Pd-C, EtOH, 23 °C; (e) EDCI•HCl,[9] HOBt, DMF, 23 °C (95%, 2 steps); (f) TMSOTf, 2,6-lutidine, CH_2Cl_2, 0 °C (100%); (g)EDCI•HCl, HOBt, Et_3N, DMF, CH_2Cl_2, 23 °C (98%).

Scheme 2



Reagents and conditions: (a) *trans*-2-methyl-2-pentenal (1.25 equiv.), Bu_2BOTf (2.0 equiv.), *i*-Pr_2NEt (1.15 equiv.), Et_2O, –100 °C → –78 °C (67%); (b) MeNH(OMe)•HCl, Me_3Al, THF, 50 °C (84%); (c) TBSCl, imidazole, DMF, 23 °C (100%); (d) DIBAL, THF, –78 °C (93%); (e) 2-methyl-1-trimethylsiloxy-1,3-butadiene, BF_3•OEt_2, CH_2Cl_2, Et_2O, –78 °C (59%); (f) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH, H_2O, 23 °C; (g) CH_2N_2, Et_2O, 0 °C (100%, 2 steps); (h) Dess–Martin periodinane, CH_2Cl_2, 23 °C (99%); (i) $NaBH_4$, MeOH, –23 °C (82%); (j) DMSO, Ac_2O, AcOH, 40 °C (74%); (k) LiOH, MeOH, H_2O, 30 °C (92%); (l) EDCI•HCl, DMAP, CH_2Cl_2, 23 °C (91%); (m) HF•pyridine, pyridine, THF, 40 °C (100%); (n) *N*-Fmoc-*N*-methyl-L-alanine, EDCI•HCl, DMAP, CH_2Cl_2, 23 °C (92%); (o) Zn, NH_4OAc, THF, H_2O, 23 °C (91%); (p) Et_2NH, MeCN, 23 °C; (q) Bop-Cl, Et_3N, CH_2Cl_2, 23 °C (32%, 2 steps); (r) $AgNO_3$, 2,6-lutidine, THF, H_2O, 70 °C (93%).

Scheme 3

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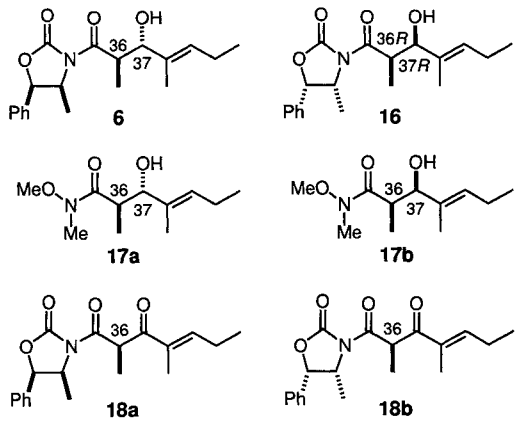
References and Notes

(1) Suenaga, K.; Mutou, T.; Shibata, T.; Itoh, T.; Kigoshi, H.; Yamada, K. *Tetrahedron Lett.* **1996**, *37*, 6771.
(2) Walker, M. A.; Heathcock, C. H. *J. Org. Chem.* **1991**, *56*, 5747. Raimundo, B. C.; Heathcock, C. H. *Synlett*, **1995**, 1213.
(3) The (36*R*,37*R*)-isomer of **6** was obtained as a minor product (14%).
(4) The stereochemistry of aldol **6** was determined as follows. Transamidation of **6** and its *syn*-diastereomer **16** that was previously[1] prepared by Evans aldol reaction gave diastereomeric amides **17a**



and **17b**, respectively. This finding indicated that the relative stereochemistry between C36 and C37 in **6** was *anti*. On the other hand, oxidation of **6** and **16** afforded diastereomeric ketones **18a** and **18b**, respectively. This fact indicated that the absolute configuration of C36 in **6** was *R*. From these results, the stereochemistry of **6** was determined to be 36*R*,37*S* (*anti*), as expected according to Heathcock and coworkers.[2]
(5) Mukaiyama, T.; Banno, K.; Narasaka, K. *J. Am. Chem. Soc.* **1974**, *96*, 7503. Paterson, I.; Smith, J. D.; Ward, R. A. *Tetrahedron*, **1995**, *51*, 9413.
(6) The stereochemistry of the C35 hydroxyl group in **8** and **10** was determined by ^1H and ^{13}C NMR analysis of the derived acetonides.[12]
(7) Dess, D. B.; Martin, J. *J. Org. Chem.* **1983**, *48*, 4155.
(8) The stereoselectivity of reduction of **9** was 1:19 (**8**:**10**). The mixture of the diastereomeric alcohols **8** and **10** was separated by column chromatography (silica gel, 1:1 hexane/ CH_2ClCH_2Cl).
(9) Sheehan, J. C.; Cruickshank, P. A.; Boshrt, G. L. *J. Org. Chem.* **1961**, *26*, 2525.
(10) Tung, R. D.; Rich, D. H. *J. Am. Chem. Soc.* **1985**, *107*, 4242.
(11) Attempts were made to construct 26-membered cyclodepsi-peptide structure, such as macrolactonization, based on other disconnections. However, they were not successful because of the low reactivity toward macrolactonization of a seco hydroxy acid or the instability of a seco amino acid.
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(13) Yamada, S.; Kasai, Y.; Shioiri, T. *Tetrahedron Lett.* **1973**, 1595.
(14) Coste, J.; Le-Nguyen, D.; Castro, B. *Tetrahedron Lett.* **1990**, *31*, 205.
(15) All new compounds gave spectroscopic data in agreement with the assigned structures. Data for the selected key compounds follow. **3**: a colorless amorphous powder; $[\alpha]_D^{27}$ +46.3 (*c* 1.41, $CHCl_3$); IR ($CHCl_3$) 3420, 3360 (br), 1755, 1675, 1630, 1515, 1465, 1390, 1140 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 0.78 (d, *J* = 6.8 Hz,

2.55 H), 0.81 (d, J = 6.8 Hz, 0.45 H), 0.85–1.07 (m, 21 H), 1.23–1.90 (m, 6 H), 2.05 (m, 1 H), 2.32 (m, 1 H), 2.89 (d, J = 4.9 Hz, 0.15 H), 2.93 (d, J = 4.9 Hz, 0.85 H), 2.96 (s, 0.45 H), 3.10 (s, 3 H), 3.12 (s, 2.55 H), 3.93 (d, J = 17.1 Hz, 0.15 H), 3.99 (d, J = 15.6 Hz, 0.85 H), 4.06 (d, J = 15.6 Hz, 0.85 H), 4.13 (dd, J = 2.0, 4.9 Hz, 1 H), 4.27 (d, J = 17.1 Hz, 0.15 H), 4.62 (d, J = 11.7 Hz, 1 H), 4.64 (dd, J = 4.9, 8.8 Hz, 0.85 H), 4.69 (dd, J = 4.9, 8.3 Hz, 0.15 H), 4.82 (m, 0.15 H), 4.85 (dd, J = 6.8, 8.8 Hz, 0.85 H), 4.90 (d, J = 11.7 Hz, 0.85 H), 4.92 (d, J = 11.7 Hz, 0.15 H), 5.29 (dd, J = 5.4, 9.3 Hz, 0.15 H), 5.47 (dd, J = 6.3, 8.3 Hz, 0.85 H), 6.74 (d, J = 8.8 Hz, 0.85 H), 6.82–6.89 (m, 0.3 H), 6.93 (d, J = 8.8 Hz, 0.85 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 11.8 (q), 12.6 (q), 17.39 [17.25] (q), 17.41 [17.7] (q), 19.0 (q), 19.5 (q), 22.0 [21.8] (q), 22.9 [23.1] (q), 24.7 [24.6] (d), 26.2 (t), 30.59 [30.65] (d), 30.74 (q), 31.1 [30.9] (d), 36.5 (q), 37.7 [37.8] (t), 38.6 (d), 51.2 [50.7] (d), 52.6 [52.4] (t), 53.7 [54.0] (d), 57.0 [57.3] (d), 73.95 [74.00] (d), 74.35 [74.40] (t), 94.4 [94.3] (s), 168.6 [168.3] (s), 170.2 [170.7] (s), 171.8 [171.6] (s), 172.3 [172.4] (s), 173.6 [173.7] (s) (the minor counter parts of doubled signals in the ratio of 5.6:1 are in brackets); MS (FAB) m/z 681 $(M + Na)^+$; HRMS (FAB) calcd for $C_{28}H_{49}{}^{35}Cl_3N_4NaO_7$ $[(M + Na)^+]$ 681.2564, found 681.2579.

4: a colorless amorphous powder; $[\alpha]_D{}^{28}$ –90.3 (c 1.09, $CHCl_3$); IR ($CHCl_3$) 3100 (br), 1685, 1645, 1460, 1290, 1250, 1105, 1055, 840 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ –0.05 (s, 3 H), 0.02 (s, 3 H), 0.72 (d, J = 6.8 Hz, 3 H), 0.89 (s, 9 H), 0.96 (t, J = 7.3 Hz, 3 H), 1.54 (br s, 3 H), 1.86 (br s, 3 H), 1.95–2.11 (m, 3 H), 2.15 (s, 3 H), 2.23–2.38 (m, 2 H), 3.67 (d, J = 9.3 Hz, 1 H), 4.16 (ddd, J = 3.4, 3.4, 8.8 Hz, 1 H), 4.53 (d, J = 11.7 Hz, 1 H), 4.63 (d, J = 11.7 Hz, 1 H), 5.30 (br t, J = 6.8 Hz, 1 H), 7.06 (m, 1 H) (signals of one proton (COOH) were not observed); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ –5.3 (q), –4.4 (q), 10.4 (q), 10.7 (q), 12.3 (q), 13.8 (q), 14.0 (q), 18.1 (s), 20.8 (t), 25.8 (q, 3 C), 28.8 (t), 38.2 (d), 73.1 (t), 75.5 (d), 80.9 (d), 127.8 (s), 130.0 (d), 134.9 (s), 143.3 (d), 173.1 (s); MS (FAB) m/z 453 $(M + Na)^+$; HRMS (FAB) calcd for $C_{22}H_{42}NaO_4SSi$ $[(M + Na)^+]$ 453.2471, found 453.2495.