Total Synthesis

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Total Synthesis of Complestatin (Chloropeptin II)**

*Zhihui Wang, Michèle Bois-Choussy, Yanxing Jia, and Jieping Zhu**

Complestatin (**1**) was first isolated in 1980 from the mycelium of *Streptomyces lavendulae* as an inhibitor of alternative pathways to the complement cascade;[1] its planar structure was elucidated by Seto and co-workers in 1989.[2] Chloropeptin I (**2**)[3] was isolated in 1994 from *Streptomyces sp*. WK-3419. The absolute configurations of the amino acid constituents of **1**[2,4] and **2** (Figure 1)[5] were elucidated through detailed NMR spectroscopy, computational, and degradation studies. Structurally, complestatin (**1**) and chloropeptin I (**2**) differ only at the position of the substituted phenyl–indole ring junction and it has been demonstrated that the former is readily isomerized to the latter under mild acidic conditions.[6] Biogenetically, both natural products are linear non-ribosomal peptides that have undergone oxidative phenolic coupling to produce the rigid cross-linked architecture.[7] Interestingly, although the shape of the rings and central strand in **1** and **2** strongly resembles that of the vancomycin family of glycopeptide antibiotics,[8] the biological activities of **1** and **2** are completely different to that of vancomycin. Indeed, both **1** and **2** are inactive against Gram-positive bacteria, but display potent activities against HIV-1-induced cytopathicity, syncyctium formation in CD-4 lymphocytes, and inhibit HIV replication by inhibition of gp 120-CD-4 binding at a low-micromolar level ($IC_{50} = 3.3$ and 2.0 μM, respectively).[4,9] It has also been demonstrated that complestatin blocks both NMDA and AMPA neurotoxicity in a noncompetitive and reversible manner. Therefore, it could also be potentially useful for preventing excitotoxicity under certain pathological conditions.[10]

The complex molecular architecture and important biological activities of **1** and **2** have attracted much attention, and have provided chemists with impetus for the development of new synthetic strategies. To develop a successful synthesis, two key issues need to be addressed: 1) epimerization during and after peptide coupling must be avoided as complestatin contains four aryl glycine units that are extremely prone to racemization, even under mild basic conditions;[11] 2) the construction of strained bismacrocycles with defined atropostereochemistry.[12] The group of Snapper and Hoveyda accomplished the first total synthesis of chloropeptin I (**2**)[13] and an atropisomer of complestatin named isocomplestatin (**3**, Figure 1).[14] These total syntheses have also allowed Snapper and Hoveyda to assign the a*R* configuration to the axial chirality of both natural products **1** and **2**. Very recently, Boger and co-workers published the first total synthesis of complestatin.[15]

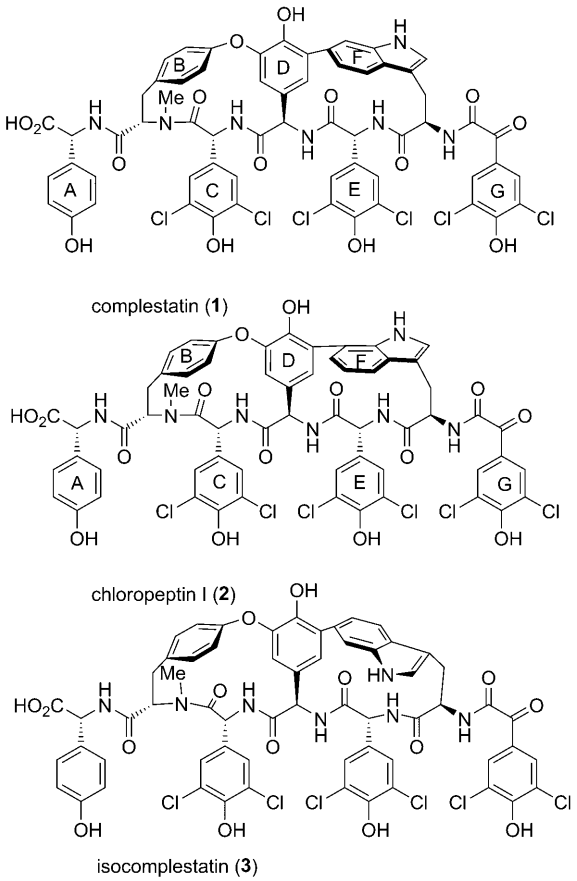

Figure 1. Structures of complestatin (**1**), chloropeptin I (**2**), and isocomplestatin (**3**).

Previous studies by our group on the synthesis of complestatin have demonstrated that the atroposelectivity of the intramolecular Suzuki–Miyaura reaction[16] for the construction of the DEFG ring is highly substrate dependent. Thus, building the DEFG ring onto the preformed ABCD macrocycle (**4**) afforded the isocomplestatin skeleton with an *aS* configuration at the biaryl axis,[14,17] whereas the cyclization of linear peptide DEFG, which was suitably functionalized to afford the fragment of complestatin (**1**), furnished the DEFG ring **5** with an *aR* configuration, identical to complestatin (**1**).[18] Based on these results, our revised synthesis

[*] Dr. Z. H. Wang, Dr. M. Bois-Choussy, Dr. Y. Jia, Dr. J. Zhu
Centre de Recherche de Gif
Institut de Chimie des Substances Naturelles, CNRS
91198 Gif-sur-Yvette Cedex (France)
Fax: (+33) 1-6907-7247
E-mail: zhu@icsn.cnrs-gif.fr
Homepage: http://www.icsn.cnrs-gif.fr/article.php3?id_article=122

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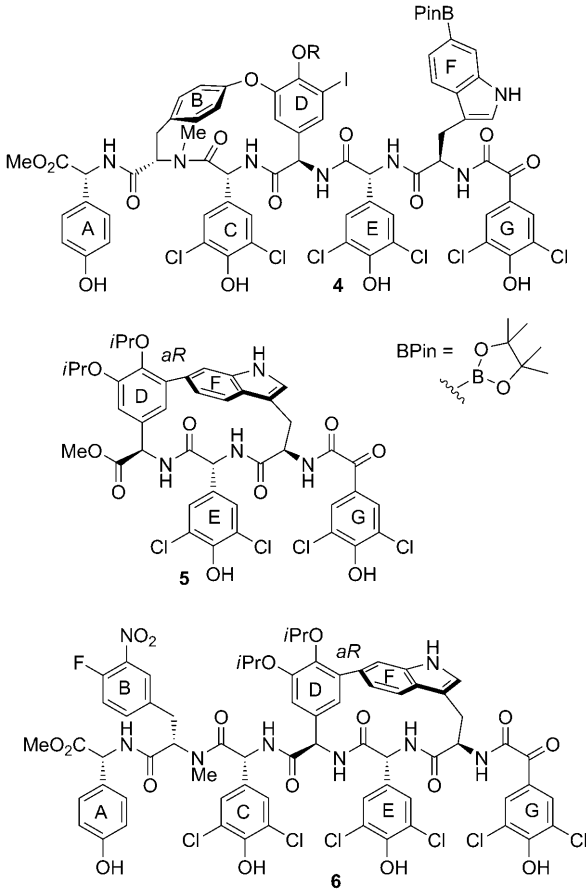
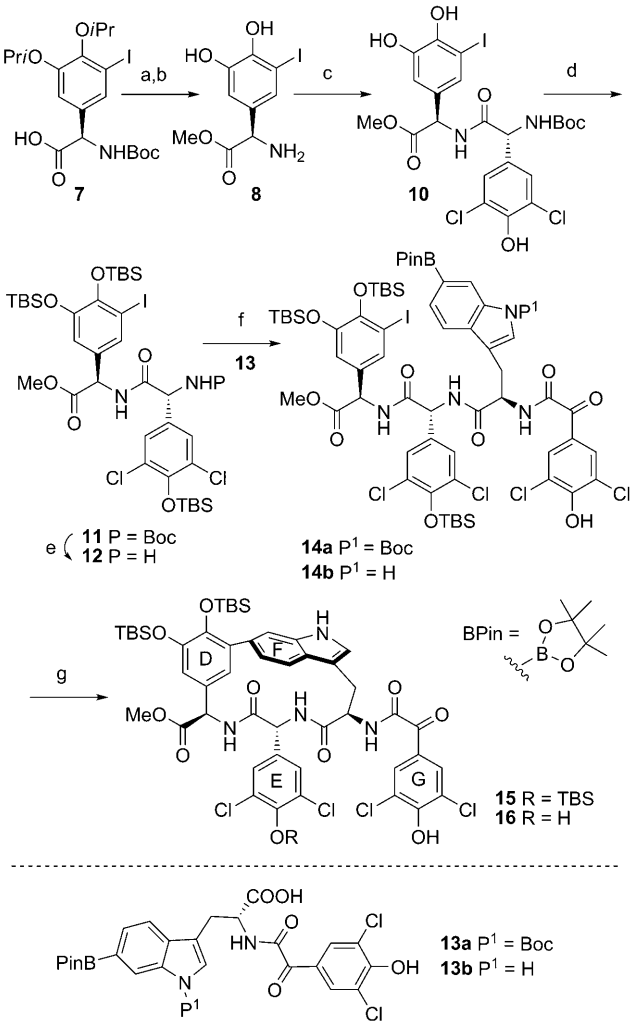
required an intramolecular Suzuki–Miyaura reaction to close the DEFG ring, followed by an intramolecular S_NAr reaction[19] to build the entire bismacrocycle of complestatin (**1**). To this end, we initially synthesized hexapeptide **6** from compound **5**.[20] However, our inability to deprotect the isopropyl ethers in **6**, as well as those in **5**, without destruction of these molecules forced us to reconsider the protective group of central amino acid D. Herein, we report the realization of a convergent total synthesis of complestatin (**1**).





Scheme 1. a) $SOCl_2$, MeOH; b) BCl_3, CH_2Cl_2; c) D-*N*-Boc-3,5-dichloro-4-hydroxyphenylglycine (**9**), DEPBT, $NaHCO_3$, THF; d) TBSCl (9 equiv), imidazole (18 equiv), DMF, 40 °C, 65% yield over 4 steps; e) 12 M HCl in MeCN (v/v = 0.07); f) HATU, 2,5-lutidine, CH_2Cl_2/THF, 80% yield; g) $[PdCl_2(dppf)]\cdot CH_2Cl_2$ (1 equiv), K_2CO_3 (10 equiv), dioxane/H_2O (15:1), 90 °C, 66% yield. HATU = *O*-(7-azabenzotriazol-1-yl)-*N*,*N*,*N*′,*N*′-tetramethyluronium hexafluorophosphate; TBS = *tert*-butyldimethylsilyl; DMF = *N*,*N*-dimethylformamide; DEPBT = 3-(diethoxyphosphoryloxy)-1,2,3-benzotriazin-4(3*H*)-one; dppf = 1,1′-bis(diphenylphosphino)ferrocene; Boc = *tert*-butyloxycarbonyl.

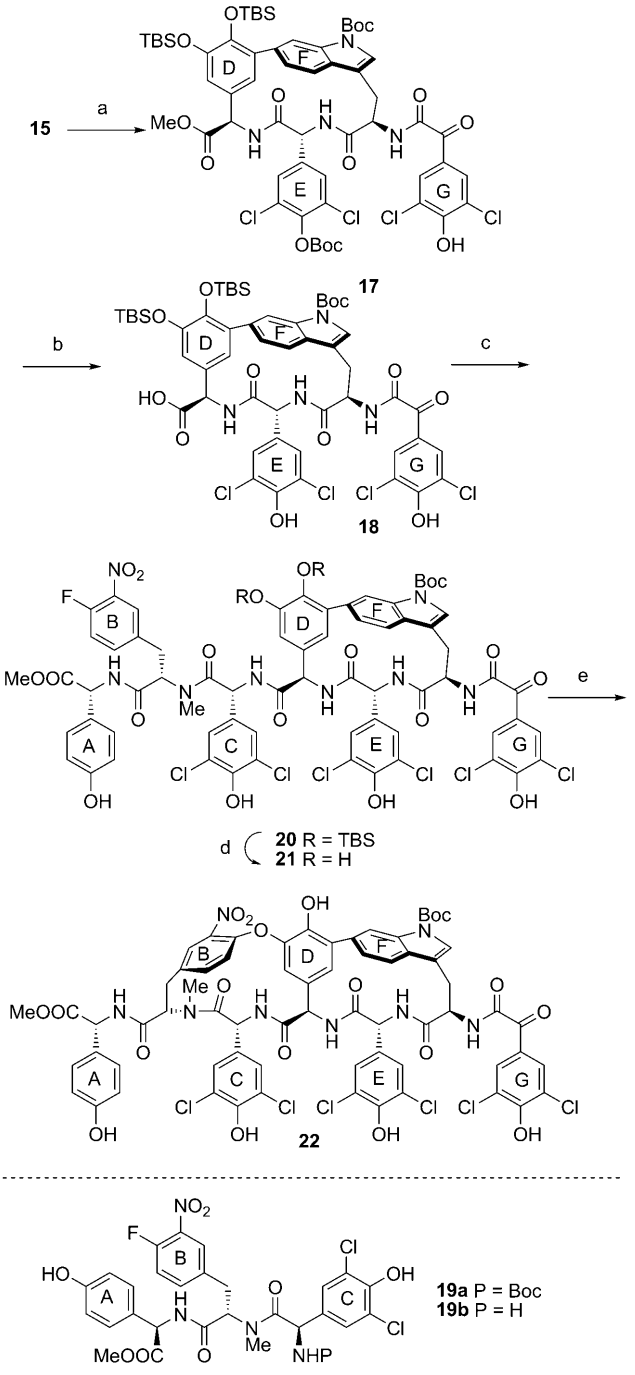
Our synthesis started from the construction of TBS-protected DEFG ring **15** (Scheme 1). Treatment of a methanol solution of D-*N*-Boc-3-iodo-4,5-diisopropyloxy phenylglycine (**7**)[18] with thionyl chloride afforded a one-pot *N*-Boc deprotection and carboxylic acid esterification to provide amino ester **8**, after a BCl_3-mediated deprotection of the isopropyl ether,[21] in quantitative yield. Coupling of **8** with D-*N*-Boc-3,5-dichloro-4-hydroxyphenylglycine (**9**) under optimized conditions (DEPBT, $NaHCO_3$, THF) furnished the dipeptide **10** in 86% yield; the three free hydroxy groups were then silylated to afford **11**. Selective removal of the *N*-Boc protecting group was realized under carefully controlled conditions using a solution of concentrated HCl in acetonitrile (v/v = 0.07) to afford **12**, which was employed directly in the following step. Other conditions including that of Sakaitani and Ohfune (TBSOTf, 2,6-lutidine, CH_2Cl_2)[22] provided a low yield of the desired product. HATU-mediated coupling of a crude sample of **12** with tryptophan derivative **13a** provided the tripeptide **14a** in 80% yield. Unfortunately, attempts to cyclize **14a** (P^1 = Boc) under a variety of conditions failed to produce the desired macrocycle. Considering that the presence of an *N*-Boc group may lead to a conformation that disfavors the ring closure, we prepared **14b** by coupling of **12** with **13b** (P^1 = H). To our delight, the intramolecular Suzuki–Miyaura reaction[23] of **14b** (P^1 = H) under our previously developed conditions ($[PdCl_2(dppf)]\cdot CH_2Cl_2$ (1.0 equiv), K_2CO_3 (10.0 equiv), dioxane/H_2O (v/v = 15:1), 90 °C) took place smoothly, in spite of the bulkiness of the TBS group, to afford the 16-membered

cyclophane **15**, together with a small amount of partially *O*-desilylated compound **16**, in 66% yield. The axial chirality of **15** was determined to be *aR* by detailed NOE studies (see the Supporting Information) as well as from the characteristically upfield shift of the α proton of amino acid F ($\delta = 4.2$ ppm). This assignment was ultimately confirmed by subsequent conversion of **15** into the natural product.

The synthesis of bicyclic compound **22** is shown in Scheme 2. Protection of the indole nitrogen in **15** [Boc_2O, DMAP, MeCN/water (v/v = 1000:1)] afforded **17**, in which the TBS ether of ring E was concurrently converted into *tert*-butoxycarbonate. The presence of a small amount of water in the reaction mixture is of utmost importance. In its absence, the reaction afforded a complex mixture of products, presumably owing to the *tert*-butoxylcarbonylation of backbone amides.[24] The lability of the TBS ether on the E ring might be due to the presence of two *ortho* chlorine atoms that caused the phenoxide to be a better leaving group. Prolonged heating of a 1,2-dichloroethane solution of **17** in the presence of trimethyltin hydroxide effected the hydrolysis of both the methyl ester and the *tert*-butoxycarbonate on the E ring to afford carboxylic acid **18** in 68% yield.[25] Coupling of **18** with tripeptide **19b**, which was obtained from the acid-mediated deprotection of **19a**, in the presence of HATU, provided the hexapeptide **20** in 60% yield. The incorporation of a base into the reaction mixture caused side reactions and should be avoided. Deprotection of the two TBS ethers was not trivial; after a number of unsuccessful trials, phenol **21** was obtained using an HBr-buffered DMF solution of potassium fluoride (pH ≈ 6). A stirred solution of the crude **21** in freshly distilled DMSO in the presence of K_2CO_3 and 4 Å molecular sieves at 30 °C provided bicyclic compound **22** in 62% overall yield from an intramolecular S_NAr reaction.[15,26] The presence of water in the reaction mixture was detrimental as it led to a complex mixture of products.

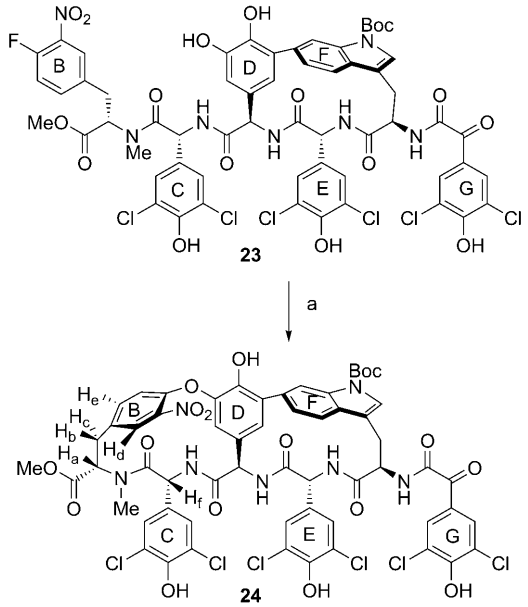
Although of no consequence, this intramolecular S_NAr reaction turned out to be highly atropostereoselective to provide a single atropisomer whose configuration was determined to be *pR* on the basis of NOE studies (see the Supporting Information). More importantly, the cyclization was regioselective as the alternative 14-membered, 17-membered, and 20-membered cyclophanes that would result from nucleophilic attack of the other hydroxy groups on rings A, C, D, and E onto the fluoro nitro aromatic system were not observed.[27] Detailed NMR spectroscopic studies indicated that the tertiary amide of residue B existed exclusively as the *cis* conformer, as is found in the natural product. Interestingly, the cyclization of pentapeptide **23**, which lacks the C-terminal 4-hydroxy phenylglycine unit afforded the *p*S atropisomer of bismacrocycle **24** (Scheme 3), wherein the tertiary amide of residue B existed preferentially as a *trans* isomer (characteristic NOE cross-peaks: H_a–H_b, H_b–H_d, H_c–H_e, H_a–N*Me*, N*Me*–H_f, $\delta_{Ha} = 4.08$ ppm versus 5.00 ppm in **22**). The crucial role of the C-terminal amino acid on the tertiary amide configuration has been noted previously by the Smith group[12e] and by ourselves.[17]

The completion of the synthesis of complestatin is detailed in Scheme 4. Reduction of the nitro group in compound **22** was problematic. After an extensive survey of

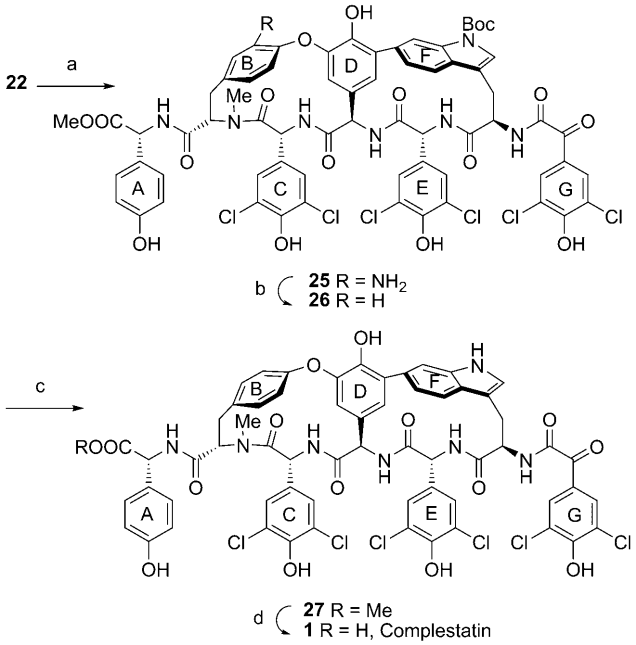


Scheme 2. a) Boc_2O (10 equiv), DMAP (0.4 equiv), MeCN/H_2O (v/v = 1000:1, *c* = 0.003 M), RT, overnight; b) Me_3SnOH, $ClCH_2CH_2Cl$, 80 °C, 3 days, 48% yield over 2 steps; c) tripeptide **19b**, HATU (1.5 equiv), THF, 60% yield; d) KF (20 equiv), conc. aqueous HBr in DMF (0.03 M, 3 equiv), RT, 2 h; e) K_2CO_3, DMSO, 4 Å M.S., 30 °C, 20 h, 62% yield over 2 steps.

reaction conditions, the reduction proceeded successfully using $SnCl_2$ as the reductant in MeOH. To guarantee a good yield of aniline **25**, the reaction had to be quenched as soon as it went to completion. We found it convenient to perform this reaction in [D_4]methanol and to monitor the progress of the reaction by ^1H NMR spectroscopy. Treatment of a THF solution of aniline **25** with *t*BuONO and H_3PO_2 afforded the



Scheme 3. a) K_2CO_3, DMSO, 4 Å M.S., 30 °C, 20 h, 60 % yield.



Scheme 4. a) $SnCl_2$, MeOH, 60 °C, 78 % yield; b) *t*BuONO, H_3PO_2, THF, 0 °C, 82 % yield; c) trifluoroethanol, microwave irradiation, 100 °C; d) LiOH, THF/H_2O, 0 °C, 83 % yield over 2 steps.

bismacrocycle **26** by in situ reduction of the diazonium salt. Removal of the *N*-Boc functional group was realized in trifluoroethanol ($pK_a \approx 12$) under microwave irradiation[28] to afford **27** which, upon saponification and purification by preparative HPLC, provided complestatin (**1**) in 83 % overall yield. The physical and spectroscopic data of the synthetic material were in accord with those described for the naturally occurring substance.

In conclusion, we have developed an efficient synthesis of complestatin (**1**). Intramolecular Suzuki–Miyaura and S_NAr reactions were employed for the construction of two macrocycles by the formation of aryl–aryl and aryl–aryl ether bonds, respectively. A [3+3] segment coupling was used for the construction of the hexapeptide backbone, which makes this synthesis highly convergent.

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