Total Syntheses of (±)-Anchinopeptolide D and (±)-Cycloanchinopeptolide D

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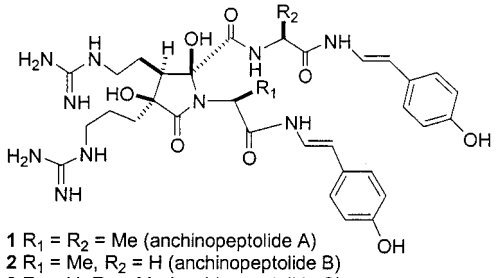
Received September 14, 1999

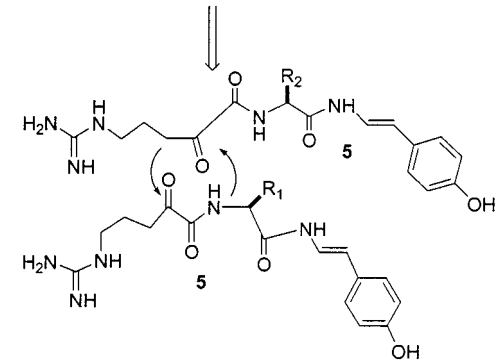
The first synthesis of (±)-anchinopeptolide D (**4**) has been accomplished in seven steps in 10% overall yield from octopamine hydrochloride (**17**), *N*-(Boc)glycine (**16**), and 5-amino-2-hydroxypentanoic acid (**22**). The key step is the aldol dimerization and hemiaminal formation of α-keto amide **26**, which gives primarily protected anchinopeptolide D **27** under kinetically controlled conditions. Cycloanchinopeptolide D (**31**) has been prepared by the unprecedented head-to-head photodimerization of the two hydroxystyrylamides of **4** using the hydrophobic effect in water to force the two side chains into close proximity so that [2 + 2] cycloaddition is faster than trans to cis double bond isomerization. Coupling of amine **21** with pyroglutamic acid affords the naturally occurring tripeptide **35**, which had been assigned glutamic acid structure **34**.

Introduction

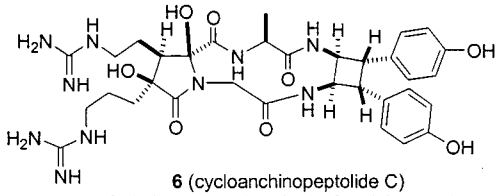
The dimeric peptide alkaloids anchinopeptolides A−D (**1**−**4**) were isolated by Minale and co-workers from the sponge *Anchinoe tenacior* collected off the coast of Tunisia.[1,2] Anchinopeptolides B−D displace specific ligands from the somatostatin receptor (62−73%), the human B2 bradykinin receptor (52−71%), and the neuropeptide Y receptor (57−80%) at 5 μg/mL.[2] These alkaloids are dimers of the modified tripeptide **5**, containing an α-keto acid derived from arginine, glycine or alanine, and hydroxystyrylamide, probably formed from tyrosine.[3] An aldol reaction will form the carbon−carbon bond. Addition of the amide to the remaining ketone will form the 5-hydroxypyrrolidinone.



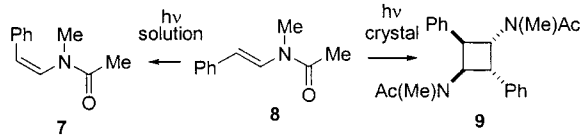
1 $R_1 = R_2$ = Me (anchinopeptolide A)
2 R_1 = Me, R_2 = H (anchinopeptolide B)
3 R_1 = H, R_2 = Me (anchinopeptolide C)
4 $R_1 = R_2$ = H (anchinopeptolide D)



They also isolated an additional alkaloid, cycloanchinopeptolide C (**6**), in which an intramolecular head-to-head [2 + 2] cycloaddition between the two hydroxystyrylamido groups resulted in the formation of a cyclobutane fused to a 12-membered ring.[2] At first glance, it appears that this cycloaddition should occur photochemically. However, enamides such as **8** undergo only trans to cis isomerization to give **7** on irradiation in solution.[4] We have recently shown that head-to-tail photodimerization occurs on irradiation of crystalline enamides in which the molecules stack in the proper orientation.[5] For instance, irradiation of crystalline **8** provides 89% of **9**.[5] In crystalline **8**, cyclobutane formation is faster than rotation about the double bond because the two molecules are held properly aligned in close proximity.



6 (cycloanchinopeptolide C)
[relative stereochemistry of cyclobutane to remainder of structure unknown]



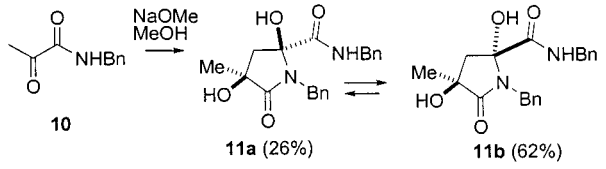
7 **8** **9**

We chose to undertake a biomimetic synthesis of anchinopeptolide D (**4**) by the aldol dimerization of **26**, a protected modified tripeptide **5** ($R_1 = R_2$ = H), to make these compounds more readily available for biological evaluation, to explore the stereochemistry of the aldol

(1) Casapullo, A.; Finamore, E.; Minale, L.; Zollo, F. *Tetrahedron Lett.* **1993**, *34*, 6297.
(2) Casapullo, A.; Minale, L.; Zollo, F.; Lavayre, J. *J. Nat. Prod.* **1994**, *57*, 1227.
(3) Schmidt, U.; Lieberknecht, A. *Angew. Chem., Int. Ed. Engl.* **1983**, *22*, 550.
(4) Hoffmann, R. W.; Eicken, K. R. *Tetrahedron Lett.* **1968**, 1759; *Chem. Ber.* **1969**, *102*, 2987.
(5) Song, F.; Snook, J. H.; Foxman, B. M.; Snider, B. B. *Tetrahedron* **1998**, *54*, 13035.

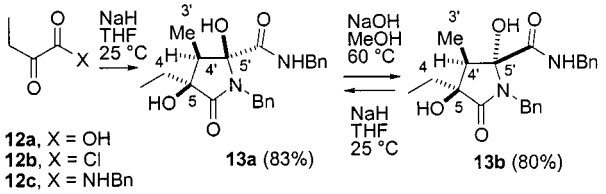
10.1021/jo991454l CCC: $19.00
Published on Web 01/14/2000

reaction and 5-hydroxypyrrolidinone formation, and to provide a substrate to explore the intramolecular [2 + 2] cycloaddition leading to cycloanchinopeptolide D. Aldol reactions of pyruvamides, which can give only a single aldol product, have been extensively explored.[6−9] The aldol adduct cyclizes to give a mixture of 5-hydroxypyrrolidinones. For instance, treatment of **10** with NaOMe in MeOH or Et_3N affords an equilibrium 30:70 mixture of **11a** and **11b**.[9] The individual isomers reequilibrate in MeOH at reflux for 1 h. Aldol dimerizations of longer chain α-keto amides, which will give mixtures of diastereomers, have not been examined. We therefore prepared *N*-benzyl-2-oxobutanamide (**12c**) to examine the stereochemistry of the aldol reaction in a simple system.

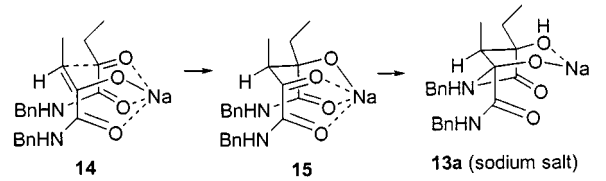


Results and Discussion

Reaction of 2-oxobutanoic acid (**12a**) with α,α-dichloromethyl methyl ether[10] gives 2-oxobutanoyl chloride (**12b**), which reacts with $BnNH_2$ and Et_3N in THF to afford 59% of *N*-benzyl-2-oxobutanamide (**12c**). We were delighted to find that treatment of **12c** with NaH in THF for 6 h at 25 °C provides 89% of a 15:1 mixture of **13a** and **13b**. The structure of **13a**, which has the same stereochemistry as the anchinopeptolides, was established by NOE studies in DMSO-d_6, in which the hydroxy and amide protons can be easily observed and assigned by HSQC and HMBC experiments. NOEs were observed between H_4 at δ 1.61 and $H_{4'}$ at δ 2.64, between C_5-OH at δ 5.15 and $H_{3'}$ at δ 0.87, between C_5-OH at δ 5.15 and $C_{5'}$-OH at δ 5.88, and between $C_{5'}$-OH at δ 5.88 and $H_{3'}$ at δ 0.87. The numbering system is based on that previously used for the anchinopeptolides.[1,2]



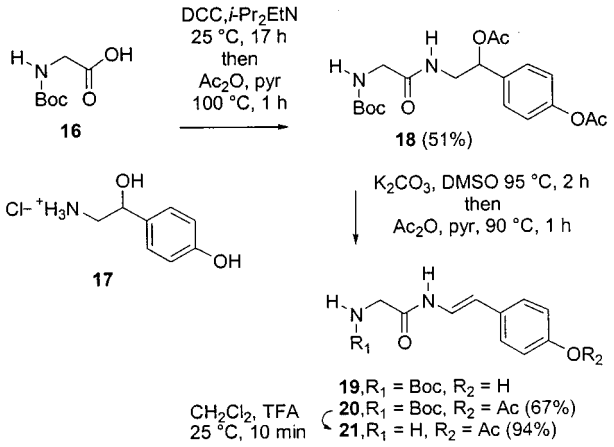
This establishes that the aldol reaction in THF occurs with the desired stereochemistry. The formation of aldol product **15** can be rationalized by consideration of a chelated transition state for the aldol reaction. Enolization should give the *Z*-enolate to avoid steric interactions between the amide and methyl groups. Transition state **14**, which leads to aldol product **15**, should be favored for the aldol reaction since the sodium can bind to all four oxygens.



Treatment of **13a** with NaOH in MeOH at reflux for 1 h results in isomerization of the 5-hydroxypyrrolidinone to give 88% of a 10:1 mixture of **13b** and **13a**. NOEs observed between H_4 at δ 1.69 and 1.51, and $H_{4'}$ at δ 2.35, between C_5−OH at δ 5.52 and $H_{3'}$ at δ 0.83, and between $H_{4'}$ at δ 2.35 and $C_{5'}$-OH at δ 7.04 established the stereochemistry of **13b**. An X-ray crystallographic structure determination confirmed the stereochemistry of **13b**.

Treatment of a solution of **13b** with NaH in THF at 25 °C for 2 h affords 75% of **13a**, 5% of **13b**, and 10% of ketoamide **12c**. This establishes that the selective formation of **13a** in THF and **13b** in MeOH is a result of a remarkable solvent effect on the stability of the hydroxypyrrolidinones rather than the kinetically favored formation of **13a** in THF. Presumably, **13a** is more stable in THF because of intramolecular hydrogen bonding between the two hydroxy groups. In the protic solvent, MeOH, intramolecular hydrogen bonding is less important and **13b** is more stable.

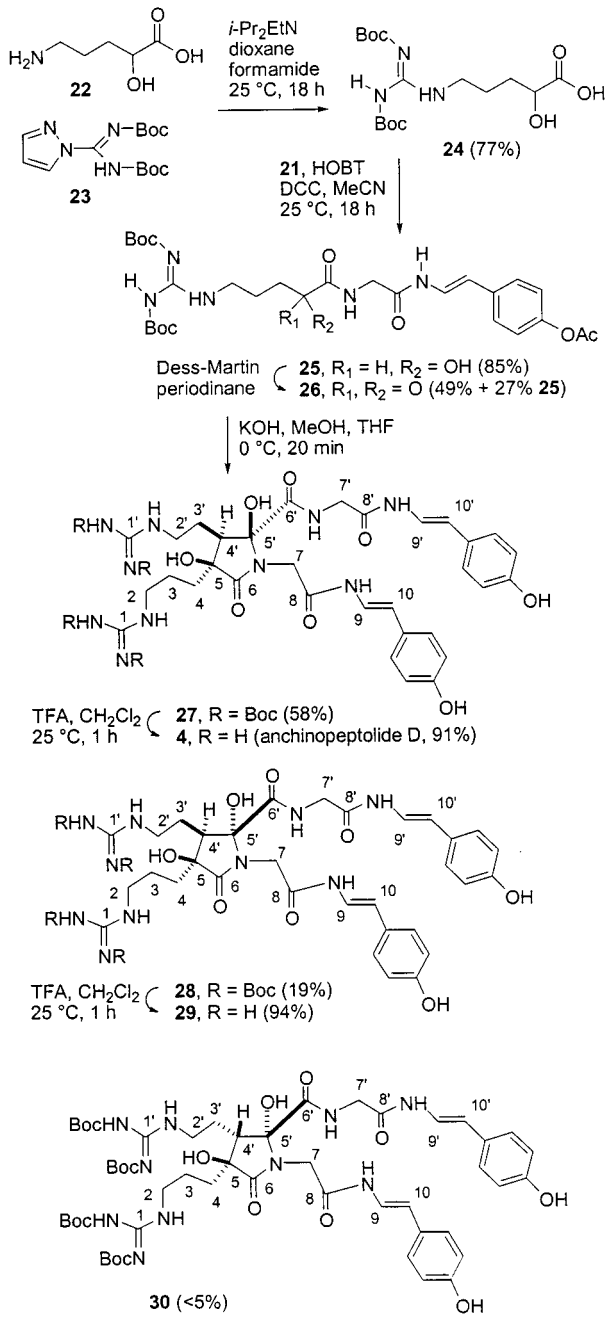
Having established that a biomimetic aldol dimerization should lead selectively to 5-hydroxypyrrolidinones with the anchinopeptolide stereochemistry, we turned our attention to the construction of the suitably protected modified tripeptide **26** to investigate the dimerization in a fully functionalized system. DCC coupling of *N*-(Boc)-glycine (**16**) with octopamine hydrochloride (**17**) and acetylation of the crude product affords 51% of diacetate amide **18**. Heating **18** with K_2CO_3 in DMSO at 90 °C[11] induces elimination of the acetate to introduce the styrene and partially hydrolyzes the aryl acetate giving a mixture of **19** and **20**. Reacetylation of the mixture affords 67% of styrylamide **20**. Removal of the Boc group in 1:1 TFA/CH_2Cl_2 at 25 °C for 10 min provides 94% of amine **21**.



Reaction of 5-amino-2-hydroxypentanoic acid (**22**)[12,13] with *N*,*N*′-bis-Boc-1-guanidylpyrazole (**23**)[14] gives 77% of the protected arginic acid derivative **24**. We were not

(6) Scudi, J. V. *J. Am. Chem. Soc.* **1937**, *59*, 1403.
(7) Pojer, P. M.; Rae, I. D. *Aust. J. Chem.* **1972**, *25*, 1737.
(8) Haeusler, J.; Schmidt, U. *Monatsch. Chem.* **1978**, *109*, 147.
(9) Stewart, K. D.; Bailey, C.; Hall, W. R.; Crouch, R. *J. Heterocycl. Chem.* **1993**, *30*, 1153.
(10) Ottenheijm, H. C. J.; Tijhuis, M. W. *Org. Synth.* **1983**, *61*, 1.
(11) Dali, H.; Sugumaran, M. *Org. Prep. Proced. Int.* **1988**, *20*, 191.
(12) Kristiansen, U.; Hedegaard, A.; Herdeis, C.; Lund, T. M.; Nielsen, B.; Hansen, J. J.; Falch, E.; Hjeds, H.; Krogsgaard-Larsen, P. *J. Neurochem.* **1992**, *58*, 1150.
(13) For another synthesis of **22**, see: Sefler, A. M.; Kozlowski, M. C.; Guo, T.; Bartlett, P. A. *J. Org. Chem.* **1997**, *62*, 93.
(14) Wu, Y.; Matsueda, G. R.; Bernatowicz, M. *Synth. Commun.* **1993**, *23*, 3055.

able to selectively protect the guanidine of arginic acid directly. Coupling of acid **24** with amine **21** using DCC and HOBT in MeCN gives 85% of hydroxy amide **25**. Oxidation of **25** with Dess−Martin periodinane in CH_2Cl_2 provides 49% of the requisite α-keto amide **26** and 27% of recovered **25**.



Treatment of α-keto amide **26** with KOH in 1:1 THF/MeOH at 0 °C for 20 min affords 58% of the desired 5-hydroxypyrrolidinone **27**, 19% of 5-hydroxypyrrolidinone **28**, with the same aldol stereochemistry, but the opposite stereochemistry at the hemiaminal center, and <5% of a third adduct **30**, derived from the other aldol adduct. Other reaction conditions, including NaH in THF, gave lower yields or less selectivity for **27**. The stereochemistry of the products was established by NOE experiments in DMSO-d_6. The protons were assigned by COSY, HSQC and HMBC experiments. In the major adduct **27**, NOEs between C_5-OH at δ 5.41, $H_{3'}$ at δ 1.78, and $C_{5'}$-OH at δ 6.63 established that the two OH groups and the guanidinoethyl chain are cis. The structure of the hydroxypyrrolidinone isomer **28** was established by an NOE between $C_{5'}$-OH at δ 7.45 and $H_{4'}$ at δ 2.42 and the absence of NOEs between C_5-OH at δ 5.34 and $H_{4'}$ at δ 2.42 and between the two hydroxy groups. The structure of the minor isomer **30** was established by NOEs between C_5-OH at δ 5.58 and $H_{4'}$ at δ 2.54 and between $C_{5'}$-OH at δ 6.86 and $H_{3'}$ at δ 1.74.

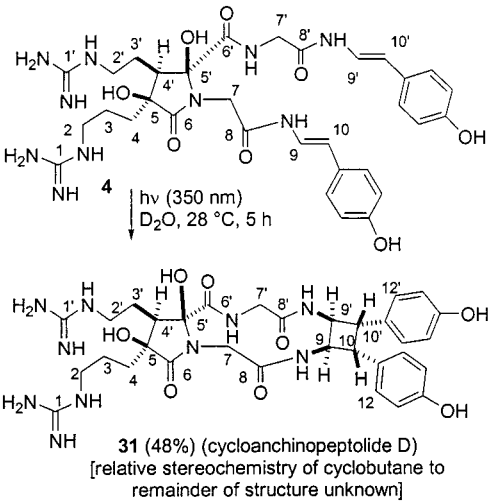
Equilibration studies confirmed these stereochemical assignments. Heating a solution of either pure **27** or **28** in CD_3OD at reflux for 1 h affords an equilibrium 2:1 mixture of **28** and **27**. This confirms the stereochemical assignments made by NOE studies, since equilibration of the hydroxypyrrolidinone isomers **27** and **28** will occur readily as observed in the equilibration of the stereoisomers of **11** and **13**. Equilibration of the aldol stereoisomers must occur more slowly than equilibration at the hemiaminal center, since opening to the acyclic keto amide, followed by enolization or a retro-aldol/aldol sequence are required to equilibrate the aldol stereoisomers. Similarly, treatment of either pure **27** or **28** with KOH in 1:1 THF/MeOH at 25 °C for 17 h provides a 6:3:1 mixture of **28**, **27**, and **30**.

The synthesis of anchinopeptolide D (**4**) was completed by cleavage of the Boc protecting groups of **27** in 1:1 TFA/CH_2Cl_2 for 1 h at 25 °C, which gives 91% of **4** as the bis trifluoroacetate salt. The ^{1}H and ^{13}C NMR in both CD_3OD and DMSO-d_6 are identical to those previously reported.[18] The stereochemistry of **4** was confirmed by NOE studies as reported for the natural product.[2] NOEs between C_5-OH at δ 5.59, $C_{5'}$-OH at δ 6.97, and $H_{3'}$ at δ 1.62 and 1.77 indicate that the two hydroxy groups and the guanidinoethyl chain are cis. Similarly, deprotection of **28** affords 94% of *epi*-anchinopeptolide D (**29**). NOEs between $C_{5'}$-OH at δ 7.44 and $H_{4'}$ at δ 2.34, but not with C_5-OH at δ 5.48 established the stereochemistry.

Synthesis of Cycloanchinopeptolide D. Our initial attempts at intramolecular photochemical [2 + 2] cycloaddition of anchinopeptolide D (**4**) were predictably disappointing. Irradiation of either **27** or the bis trifluoroacetate salt of **4** in CD_3OD at 350 nm for 4 h results in trans to cis isomerization of the double bonds as expected based on solution studies of other enamides.[4,5] We hypothesized that the choice of solvent should have a profound effect on the photochemistry. The bis trifluoroacetate salt of **4** should be soluble in water because it is a dication. In water, the hydrophobic effect should cause the two hydroxystyrylamido groups to pack closely together to minimize repulsive interactions between water and the two nonpolar side chains. If the orientation of the side chains is appropriate, [2 + 2] cycloaddition could be faster than trans to cis isomerization of the double bond. The hydrophobic effect has been shown to lead to different products from irradiation of stilbenes and alkyl cinnamates in water and nonpolar solvents.[19−22]

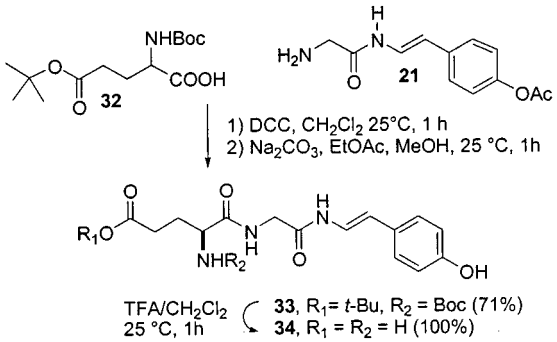
(15) Hagihara, M.; Schreiber, S. L. *J. Am. Chem. Soc.* **1992**, *114*, 6570.
(16) Bastiaans, H. M. M.; van der Baan, J. L.; Ottenheijm, H. C. J. *Tetrahedron Lett.* **1995**, *36*, 5963.
(17) Bernatowicz, M.; Wu, Y.; Matsueda, G. R. *Tetrahedron Lett.* **1993**, *34*, 3389.
(18) Our ^{13}C NMR data for **4**, which were referenced to the central peak of CD_3OD at δ 49.15, are 0.3 ppm downfield from the literature data, which are referenced to δ 48.85. The coupling constants for $H_{4'}$ are 4.8 and 10 Hz. $C_{5'}$ absorbs at δ 91.0 not 90.1 as reported. Dr. Agostino Casapullo, private communication, January 1999.

We were delighted to find that irradiation of a 0.005 M solution of the bis trifluoroacetate salt of **4** in D_2O at 350 nm at 28 °C for 5 h provides 48% of cycloanchinopeptolide D (**31**). In CD_3OD, the cyclobutane protons, H_9 at δ 4.56, $H_{9'}$ at δ 4.83, H_{10} at δ 4.11, and $H_{10'}$ at δ 4.00, were assigned by COSY, HSQC, and HMBC experiments. NOEs between $H_{9'}$ at δ 4.83 and $H_{12'}$ at δ 6.79, between H_9 at δ 4.56 and H_{12} at δ 6.72, and between $H_{9'}$ at δ 4.83 and H_9 at δ 4.56, establish that the cyclobutane was formed by head-to-head dimerization of trans hydroxystyrylamides. The stereochemistry of the pyrrolidinone of **31** was assumed to be the same as that of **4**. The relative stereochemistry between the pyrrolidinone and the cyclobutane was not determined, as was the case in the structure determination of cycloanchinopeptolide C (**6**).[2]



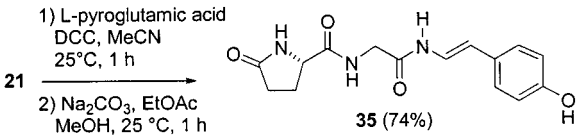
The spectral data for cycloanchinopeptolide D (**31**) are very similar to those reported for cycloanchinopeptolide C (**6**). The geminal coupling constant for the C_7 methylene protons of **31**, 17.0 Hz, is very close to that observed for glycine in proteins and in **6**. The geminal coupling constant for the $C_{7'}$ methylene protons, 13.4 Hz, is much smaller than typical for glycine and initially caused some concern. In **6**, this coupling constant is not observed since an alanine rather than glycine is present in this chain. Barfield suggested that the geminal coupling constants in the glycine residues of a peptide backbone depend on both dihedral angles ϕ and θ in the backbone. The variation of the coupling constants was calculated to be as much as 8 Hz.[23,24] Molecular mechanics calculations indicate that **31** exists in one highly preferred conformation with ϕ and θ values that should result in a small coupling constant.

Synthesis of Tripeptides 34 and 35. Minale and coworkers also reported the isolation of 5.6 mg of tripeptide **34** from the same sponge that produced the anchinopeptolides.[25] The structure was established by NMR spectroscopic analysis. Hydrolysis with 6 N HCl, reaction with Marfey's reagent, and HPLC analysis established the L-glutamic acid configuration. We chose to prepare this tripeptide since acetoxystyrylglycine (**21**) was available from our anchinopeptolide D synthesis. DCC coupling of protected glutamic acid **32** with **21** followed by hydrolysis of the acetate with sodium carbonate affords 71% of **33**. Cleavage of the Boc groups with 1:1 TFA/CH_2Cl_2 provides the trifluoroacetate salt of **34**.



The ^1H and ^{13}C NMR spectra of **34** are very different than those reported for the natural product. The NMR spectra of amino acids are very sensitive to pH. We therefore examined the spectra of **34** in basic, neutral and acidic CD_3OD. The spectral data of the natural product do not match well with those of the synthetic material obtained at any pH. The carboxylic acid carbon at δ 170.4 does not match that reported at δ 181.5. The C_2 methylene group triplet at δ 2.56 does not match the reported multiplets at δ 2.35 and 2.55 for this methylene group.

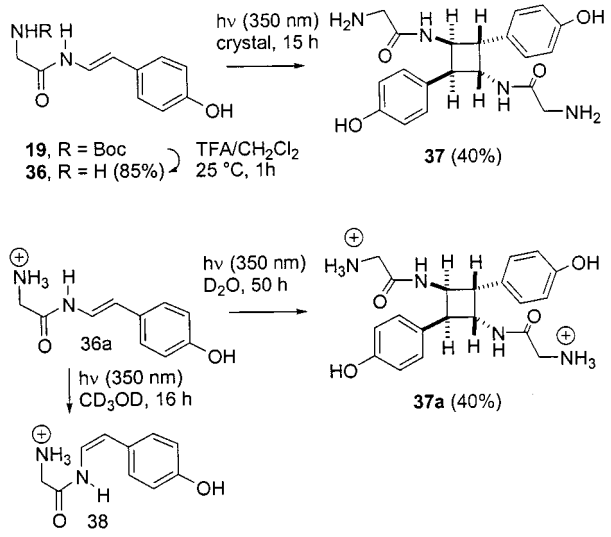
The very different chemical shifts of the C_2 methylene protons of the natural product suggested that this carbon is in a ring. Tripeptide **35** with a pyroglutamate should have proton and carbon shifts similar to those reported and will give L-glutamic acid on hydrolysis. The failure to observe a parent ion at *m*/*z* 321 with either EI or FAB ionization is also consistent with **35** which could give a parent ion at *m*/*z* 303 that could be misinterpreted as loss of water from **34** in the mass spectrometer.



We therefore prepared **35** in 74% yield by treatment of **21** with L-pyroglutamic acid and DCC in acetonitrile and hydrolysis of the acetate with Na_2CO_3. The ^1H and ^{13}C NMR spectral data for **35** are identical to those reported for the natural product[18] indicating that the structure of the natural tripeptide should be revised from **34** to **35**. The optical rotation of synthetic **35**, $[\alpha]^{25}_D$ +2.9, is very different from the reported value for the tripeptide, $[\alpha]^{25}_D$ −4.6. We are unable to comment on the significance of this discrepancy, except to note that only 5.6 mg of natural tripeptide was isolated and the sign of optical rotation is very sensitive to trace impurities because the magnitude is so small.

(19) Devanathan, S.; Ramamurthy, V. *J. Photochem. Photobiol. A* **1987**, *40*, 67.
(20) Syamala, M. S.; Ramamurthy, V. *J. Org. Chem.* **1986**, *51*, 3712.
(21) Ito, Y.; Kajita, T.; Kunimoto, K.; Matsuura, T. *J. Org. Chem.* **1989**, *54*, 587.
(22) Li, Y.; Deng, X. H.; Wang, X. H.; Tung, C. H. *Chin. Chem. Lett.* **1994**, *5*, 287.
(23) Barfield, M.; Hruby, V. J.; Meraldi, J. P. *J. Am. Chem. Soc.* **1976**, *98*, 1308.
(24) Bystrov, V. F. *Prog. Nucl. Magn. Reson. Spectrosc.* **1976**, *10*, 41.
(25) Casapullo, A.; Minale, L.; Zollo, F. *Tetrahedron Lett.* **1994**, *35*, 2421.

Photochemistry of Amine 36 and Ammonium Salt 36a. We briefly examined the photochemistry of hydroxystyrylglycine **36**. Cleavage of the Boc group of **19** with TFA/CH_2Cl_2 affords 85% of **36**. Irradiation of **36** at 350 nm in CD_3OD results only in trans to cis isomerization. No reaction occurs on irradiation of crystalline **36**. Rapid evaporation of a solution of **36** in MeOH affords a thin film that gives 40% of head-to-tail dimer **37** on irradiation for 16 h at 350 nm. The spectral data of **37** are similar to those of analogous cyclobutanes such as **9** that we have previously prepared by solid-state photodimerization.[5] Presumably, rapid evaporation of the MeOH solution of **36** affords a phase with closely packed and properly oriented hydroxystyrylamido residues that undergoes [2 + 2] cycloaddition on irradiation. The different phase formed on slow evaporation does not have properly oriented hydroxystyrylamido residues and cannot undergo either cycloaddition or trans to cis double bond isomerization on irradiation.



Since we have shown that use of water as the solvent has a profound effect on the photochemistry of the dication anchinopeptolide D (**4**), we examined the photochemistry of **36a**, the trifluoroacetate salt of **36**, in both water and MeOH. Irradiation in CD_3OD for 16 h provides a mixture of the **36a**, the cis isomer, and decomposition products. Irradiation for 30 h affords only decomposition products. On the other hand, irradiation of **36a** in D_2O for 50 h affords 40% of the dimeric dication **37a**, which provides diamine **37** on neutralization. This suggests that ammonium salt **36a** aggregates in water due to the hydrophobic effect to place the enamide double bonds of two molecules in close proximity in a head-to-tail orientation so that cycloaddition is faster than trans to cis isomerization.

In conclusion, we have completed the first synthesis of (±)-anchinopeptolide D (**4**), which proceeds in seven steps in 10% overall yield from octopamine hydrochloride (**17**), *N*-(Boc)glycine (**16**), and 5-amino-2-hydroxypentanoic acid (**22**). We have shown that the aldol dimerization and hemiaminal formation of α-keto amide **12c** gives primarily diastereomer **13a** with NaH in THF. In basic MeOH, equilibration of the hemiaminal center affords mainly **13b**, which can be reconverted to **13a** with NaH in THF. Cycloanchinopeptolide D (**31**) has been prepared by the unprecedented head-to-head photodimerization of the two hydroxystyrylamides of **4** using the hydrophobic effect in water to force the two side chains into close proximity so that [2 + 2] cycloaddition is faster than trans to cis double bond isomerization. Coupling of amine **21** with pyroglutamic acid affords the naturally occurring tripeptide **35**, which had been incorrectly assigned glutamic acid structure **34**.

Experimental Section

General Methods. NMR spectra were recorded at 400 MHz in $CDCl_3$, CD_3OD, or DMSO-d_6 as indicated. Negative NOEs were observed for **4** and **27**−**31** in DMSO-d_6. Positive NOEs were observed for all compounds in CD_3OD and for **13** in DMSO-d_6. Chemical shifts are reported in δ and coupling constants in Hz. IR spectra are reported in cm^{-1}. Reversed-phase chromatography was carried out on J. T. Baker Bakerbond Octadecyl (C_{18}) 40 μm prep LC packing.

2-Oxo-*N*-(phenylmethyl)butanamide (12c). A mixture of 2-oxobutanoic acid (1.021 g, 10 mmol) and α,α-dichloromethyl methyl ether (0.91 mL, 10 mmol) was heated at 50 °C for 30 min in a flask that was connected to an open $CaCl_2$ drying tube. The residue was dissolved in 5 mL of THF, and the acid chloride solution was added to a solution of benzylamine (1.09 mL, 10 mmol) and Et_3N (1.40 mL, 10 mmol) in 15 mL of THF at 0 °C over 30 min with rapid stirring. The solid was filtered off, and water was added to the filtrate. The solution was extracted with three portions of EtOAc. The combined extracts were dried over Na_2SO_4. The solvent was removed and the residue was purified on silica gel (3:1 CH_2Cl_2/hexane) to give 1.128 g (59%) of **12c**: mp 79−80 °C; ^{1}H NMR ($CDCl_3$) 7.25−7.37 (m, 5), 7.28 (br, 1, NH), 4.47 (d, 2, J = 6.1), 2.98 (q, 2, J = 7.2), 1.11 (t, 3, J = 7.2); ^{13}C NMR ($CDCl_3$) 199.5, 159.9, 137.0, 128.8 (2 C), 127.84 (2 C), 127.80, 43.3, 30.3, 7.0; IR (KBr) 3253, 1723, 1671.

(2α,3α,4α)-4-Ethyl-2,4-dihydroxy-3-methyl-5-oxo-*N*,1-bis(phenylmethyl)-2-pyrrolidinecarboxamide (13a). To a solution of **12c** (191 mg, 1 mmol) in 5 mL of THF at 25 °C, was added a suspension of NaH (60% w/w in mineral oil) (80 mg, 2 mmol) in 5 mL of THF over 2 h with rapid stirring. After addition, the mixture was stirred for 4 h and neutralized with 0.1 N HCl. The mixture was concentrated under reduced pressure, and the residue was extracted with four portions of $CHCl_3$. The combined extracts were dried over $MgSO_4$ and concentrated to give a 15:1 mixture of **13a** and **13b**. The mixture was purified on silica gel (3:1 CH_2Cl_2/EtOAc) to give 159 mg (83%) of **13a**: mp 164−165 °C; ^{1}H NMR (DMSO-d_6) 8.69 (t, 1, J = 6.1, NH), 7.16−7.29 (m, 10), 5.88 (s, 1, OH), 5.15 (s, 1, OH), 4.33 (d, 1, J = 15.9), 4.22 (dd, 1, J = 6.1, 14.7), 4.17 (dd, 1, J = 6.1, 14.7), 4.12 (d, 1, J = 15.9), 2.64 (q, 1, J = 7.3), 1.61 (q, 2, J = 7.3), 0.87 (d, 3, J = 7.3), 0.82 (t, 3, J = 7.3); ^{13}C NMR (DMSO-d_6) 175.7, 169.4, 139.3, 137.6, 128.2 (2 C), 127.8 (2 C), 127.6 (2 C), 127.1 (2 C), 126.7, 126.5, 90.5, 75.2, 43.7, 42.6, 41.3, 28.7, 8.2, 7.5; IR (KBr) 3370, 1685.

(2α,3β,4β)-4-Ethyl-2,4-dihydroxy-3-methyl-5-oxo-*N*,1-bis(phenylmethyl)-2-pyrrolidinecarboxamide (13b). A solution of **13a** (18.3 mg, 0.048 mmol) in 1 mL of CH_3OH was treated with NaOH (2 mg, 0.05 mmol). The mixture was heated at 60 °C for 1 h. The solvent was removed, and H_2O was added. The mixture was extracted with 4 portions of $CHCl_3$. The combined extracts were dried over $MgSO_4$ and concentrated to give a 1:10 mixture of **13a** and **13b**. The residue was purified on silica gel (3:1 CH_2Cl_2/EtOAc) to give 14.7 mg (80%) of **13b**: mp 159−161 °C; ^{1}H NMR (DMSO-d_6) 9.03 (t, 1, J = 6.1, NH), 7.14−7.31 (m, 10), 7.04 (s, 1, OH), 5.52 (s, 1, OH), 4.30 (d, 1, J = 15.2), 4.17 (dd, 1, J = 6.1, 15.2), 4.16 (d, 1, J = 15.2), 4.11 (dd, 1, J = 6.1, 15.2), 2.35 (q, 1, J = 7.3), 1.69 (dq, 1, J = 7.3, 14.6), 1.51 (dq, 1, J = 7.3, 14.6) 0.83 (d, 3, J = 7.3), 0.82 (t, 3, J = 7.3); ^{13}C NMR (DMSO-d_6) 174.1, 171.1, 138.6, 137.5, 128.2 (2 C), 127.9 (2 C), 127.7 (2 C), 127.2 (2 C), 126.9, 126.7, 91.0, 76.6, 44.7, 42.6, 42.4, 26.2, 8.5, 6.3; ^{1}H NMR (CD_3OD) 7.16−7.32 (m, 10), 4.46 (d, 1, J = 15.2), 4.34 (d, 1, J = 15.2), 4.17 (d, 1, J = 14.8), 4.02 (d, 1, J = 14.8), 2.34 (q, 1, J = 7.2), 1.88 (qd, 1, J = 7.3, 15.2), 1.65 (qd, 1, J = 7.3,

15.2), 0.93 (d, 3, $J = 7.2$), 0.90 (t, 3, $J = 7.3$); ^{13}C NMR (CD_3OD) 176.9, 172.7, 139.5, 138.2, 129.8 (2 C), 129.7 (2 C), 129.4 (2 C), 128.8 (2 C), 128.5, 128.5, 92.7, 79.0, 46.8, 44.4, 44.0, 27.4, 9.0, 6.7; IR (KBr) 3309, 1685.

Reequilibration of 13b in THF with NaH. A solution of **13b** (16 mg, 0.047 mmol) in 2 mL of THF was treated with NaH (60% w/w in mineral oil) (7 mg, 0.175 mmol). The solution was stirred at 25 °C for 2 h and neutralized with 0.1 N HCl. The solution was extracted with three portions of EtOAc. The combined extracts were dried over $MgSO_4$ and concentrated to give 15 mg of crude product. Analysis of the NMR spectrum indicated that the mixture contained 75% of **13a**, 5% of **13b**, 10% of ketoamide **12c**, and 10% of uncharacterized material.

***N*-[2-(4-Acetoxyphenyl)acetoxyethyl](Boc)glycinamide (18).** To a mixture of *N*-(Boc)glycine (**16**) (1.752 g, 10 mmol) and DCC (2.269 g, 11 mmol) in 16 mL of CH_2Cl_2 were added octopamine hydrochloride (**17**) (1.896 g, 10 mmol) and *N*,*N*-diisopropylethylamine (1.75 mL, 10 mmol) in 8 mL of DMF. The reaction mixture was stirred at 25 °C for 17 h, filtered, and concentrated under reduced pressure to afford a gummy oil. Acetic anhydride (10 mL) and pyridine (3 mL) were added, and the mixture was heated at 100 °C under N_2 for 1 h. The mixture was cooled and poured onto ice, and the resulting solution was extracted with three portions of EtOAc. The combined organic extracts were washed with water and brine and dried over $MgSO_4$. The solvent was removed under reduced pressure and the residue was purified on silica gel (2:1 EtOAc/CH_2Cl_2) to give 2.032 g (51%) of **18**: mp 114−116 °C; ^{1}H NMR ($CDCl_3$) 7.36 (d, 2, $J = 8.8$), 7.09 (d, 2, $J = 8.8$), 6.38 (br, 1, NH), 5.85 (dd, 1, $J = 4.4, 8.0$), 5.09 (br, 1, NH), 3.75 (d, 2, $J = 6.0$), 3.71 (ddd, 1, $J = 4.4, 6.0, 14.0$), 3.58 (ddd, 1, $J = 5.4, 8.0, 14.0$), 2.30 (s, 3), 2.10 (s, 3), 1.46 (s, 9); ^{13}C NMR ($CDCl_3$) 170.2, 169.6, 169.3, 156.0, 150.7, 135.1, 127.6 (2 C), 121.8 (2 C), 80.4, 73.7, 44.5, 44.1, 28.3 (3 C), 21.1, 21.1; IR (KBr) 3392 (br), 1750, 1719, 1685.

(*E*)-*N*-[2-(4-Acetoxyphenyl)ethenyl](Boc)glycinamide (20). A mixture of **18** (2.00 g, 5.07 mmol) and K_2CO_3 (1.68 g, 1.22 mmol) in 13 mL of DMSO was heated at 95 °C under N_2 for 2 h. The mixture was cooled to 25 °C and poured onto ice. The resulting solution was extracted with six portions of EtOAc. The combined organic extracts were washed with brine and dried over $MgSO_4$. The solvent was removed under reduced pressure and the residue was purified on silica (3:1 CH_2Cl_2/EtOAc) to give 0.618 g (36%) of **20**, followed by 0.070 g of a mixture of **20** and **19**, and 0.508 g of **19**.

Data for **19**: mp 187 °C dec; ^{1}H NMR (CD_3OD) 7.26 (d, 1, $J = 14.8$), 7.15 (d, 2, $J = 8.8$), 6.71 (d, 2, $J = 8.8$), 6.19 (d, 1, $J = 14.8$), 3.79 (s, 2), 1.46 (s, 9); ^{13}C NMR 170.0, 158.6, 157.7, 129.2, 127.9 (2 C), 121.3, 116.6 (2 C), 115.4, 80.9, 44.6, 28.8 (3 C); IR (KBr) 3298, 1681, 1654.

Data for **20**: mp 138−140 °C; ^{1}H NMR ($CDCl_3$) 8.18 (br, 1, NH), 7.42 (dd, 1, $J = 11.2, 14.8$), 7.31 (d, 2, $J = 8.8$), 7.01 (d, 2, $J = 8.8$), 6.13 (d, 1, $J = 14.8$), 5.21 (br, 1, NH), 3.90 (d, 2, $J = 6$), 2.29 (s, 3), 1.49 (s, 9); ^{13}C NMR ($CDCl_3$) 169.8, 167.4, 156.6, 149.5, 134.0, 126.6 (2 C), 122.5, 121.9 (2C), 113.0, 80.7, 44.6, 28.5 (3 C), 21.3; IR (KBr) 3334, 1698, 1677, 1655.

The mixture of **20** and **19** and pure **19** were dissolved in 5 mL of acetic anhydride and 0.20 mL of pyridine. The solution was heated at 90 °C for 1 h. The reaction solution was cooled to 25 °C and poured onto ice. The mixture was extracted with six portions of EtOAc. The combined extracts were washed with water and brine, and dried over $MgSO_4$. The solvent was removed under reduced pressure and the residue was purified on silica gel (3:1 CH_2Cl_2/EtOAc) to give an additional 0.516 g (31%) of **20**.

(*E*)-*N*-[2-(4-Acetoxyphenyl)ethenyl]glycinamide (21). Enamide **20** (820 mg, 2.46 mmol) was dissolved in 5 mL of CH_2Cl_2 and 5 mL of trifluoroacetic acid and the solution was stirred at 25 °C for 10 min. The solvent was removed under reduced pressure. EtOAc (30 mL) was added to the residue and saturated Na_2CO_3 aqueous solution was added until the pH was 8. The solution was filtered and the filtrate was extracted with four portions of EtOAc. The combined extracts were dried over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was purified on silica gel (2:1 $CHCl_3$/MeOH) to give 540 mg (94%) of **21**: mp 136−137.5 °C; ^{1}H NMR (CD_3OD) 7.45 (d, 1, $J = 14.8$), 7.35 (d, 2, $J = 8.8$), 7.01 (d, 2, $J = 8.8$), 6.24 (d, 1, $J = 14.8$), 3.36 (s, 2), 2.26 (s, 3); ^{13}C NMR (CD_3OD) 172.8, 171.4, 151.0, 135.8, 127.4 (2 C), 123.9, 123.1 (2 C), 113.8, 45.1, 21.1; IR (KBr) 3390, 3274, 1750, 1645. Anal. Calcd for $C_{12}H_{14}N_2O_3$: C, 61.53; H, 6.02; N, 11.96. Found: C, 61.38; H, 5.95; N, 11.85.

5-[[Bis[[(1,1-dimethoxy)carbonyl]amino]methylene]-amino]-2-hydroxypentanoic Acid (24). To a mixture of 5-amino-2-hydroxypentanoic acid (**22**)[12,13] (148 mg, 1.11 mmol) and diisopropylethylamine (446 μL, 2.56 mmol) in 3 mL of formamide was added *N*,*N*′-bis-Boc-1-guanidylpyrazole (**23**)[14] (480 mg, 1.53 mmol) in 1.5 mL of 1,4-dioxane dropwise. The reaction mixture was stirred at 25 °C for 18 h, 9 mL of 1 M HCl was added, and the mixture was extracted with six portions of EtOAc. The combined EtOAc extracts were washed with brine and dried over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was purified on silica gel (15:1 CH_2Cl_2/MeOH containing 0.1% formic acid) to give 0.321 g (77%) of **24**: mp 84−85 °C; ^{1}H NMR (CD_3OD) 4.16 (dd, 1, $J = 3.6, 7.2$), 3.41 (t, 2, $J = 6.8$), 1.85 (m, 1), 1.72 (m, 3), 1.53 (s, 9), 1.48 (s, 9); ^{13}C NMR (CD_3OD) 177.9, 164.5, 157.7, 154.3, 84.6, 80.7, 71.3, 41.6, 32.6, 28.7 (3 C), 28.4 (3 C), 26.3; IR (KBr) 3333, 1723, 1641, 1618.

(*E*)-5-[[Bis[[(1,1-dimethoxy)carbonyl]amino]methylene]-amino]-2-hydroxypentanoyl-*N*-[2-(4-acetoxyphenyl)ethenyl]glycinamide (25). A mixture of **24** (180 mg, 0.48 mmol), **21** (113 mg, 0.48 mmol), DCC (109 mg, 0.53 mmol), and HOBt (65 mg, 0.48 mmol) in 5 mL of CH_3CN was stirred at 25 °C for 12 h. The mixture was filtered, and the filtrate was evaporated to dryness. The residue was purified on silica gel (3:1 EtOAc/CH_2Cl_2) to give 241 mg (85%) of **25**: ^{1}H NMR ($CDCl_3$) 11.47 (s, 1, NH), 8.91 (d, 1, $J = 10.8$, NH), 8.60 (t, 1, $J = 6.0$, NH), 7.82 (t, 1, $J = 6.0$, NH), 7.40 (dd, 1, $J = 10.8, 14.8$), 7.27 (d, 2, $J = 8.8$), 6.98 (d, 2, $J = 8.8$), 6.08 (d, 1, $J = 14.8$), 6.07 (s, 1, OH), 4.28 (br, 1), 4.09 (dd, 1, $J = 6.0, 16.1$), 4.01 (dd, 1, $J = 6.0, 16.1$), 3.62 (m, 1), 3.38 (m, 1), 2.29 (s, 3), 2.00 (m, 1), 1.76 (m, 3), 1.49 (s, 18); ^{13}C NMR ($CDCl_3$) 176.2, 169.5, 166.6, 162.7, 156.9, 153.1, 149.2, 133.8, 126.4 (2 C), 122.4, 121.7 (2 C), 112.7, 83.6, 79.9, 72.6, 43.4, 39.9, 29.2, 28.2 (3 C), 28.0 (3 C), 26.5, 21.1; IR (KBr) 3330, 1718, 1654, 1612. Anal. Calcd for $C_{28}H_{41}N_5O_9$: C, 56.84; H, 6.98; N, 11.84. Found: C, 56.07; H, 6.44; N, 12.42.

(*E*)-5-[[Bis[[(1,1-dimethoxy)carbonyl]amino]methylene]-amino]-2-oxpentanoyl-*N*-[2-(4-acetoxyphenyl)ethenyl]-glycinamide (26). To a solution of **25** (110 mg, 0.186 mmol) in 5 mL of CH_2Cl_2 at 0 °C was added Dess−Martin periodane (87 mg, 0.21 mmol). The mixture was stirred at 0 °C for 30 min. A 1:1 (v/v) mixture of saturated $NaHCO_3$ and 10% $Na_2S_2O_3$ solution was added. The resulting mixture was extracted with three portions of EtOAc. The combined extracts were dried over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was purified on silica gel (2:1 CH_2Cl_2/EtOAc) to give 30 mg (27%) of recovered **25** preceded by 54 mg (49%) of **26**: mp 160 °C dec; ^{1}H NMR ($CDCl_3$) 11.47 (s, 1, NH), 8.52 (d, 1, $J = 10$, NH), 8.41 (t, 1, $J = 5.4$, NH), 7.70 (t, 1, $J = 5.9$, NH), 7.40 (dd, 1, $J = 10.8, 14.8$), 7.29 (d, 2, $J = 8.8$), 7.00 (d, 2, $J = 8.8$), 6.12 (d, 1, $J = 14.8$), 4.08 (d, 2, $J = 6.0$), 3.43 (dt, 2, $J = 5.9, 7.1$), 2.99 (t, 2, $J = 7.1$), 2.30 (s, 3), 1.92 (tt, 2, $J = 7.1, 7.1$), 1.50 (s, 9), 1.49 (s, 9); ^{13}C NMR ($CDCl_3$) 197.2, 169.6, 165.4, 163.3, 160.6, 156.4, 153.2, 149.4, 133.6, 126.6 (2 C), 122.1, 121.8 (2 C), 113.2, 83.3, 79.6, 42.9, 39.8, 34.2, 28.3 (3 C), 28.0 (3 C), 22.7, 21.1; IR (KBr) 3326, 1751, 1725, 1639. Anal. Calcd for $C_{28}H_{39}N_5O_9$: C, 57.03; H, 6.67; N, 11.88. Found: C, 56.98; H, 6.37; N, 12.03.

(2α,3α,4α)-, (2α,3*β*,4*β*)-, and (2α,3α,4*β*)-3-[2-[[Bis[[(1,1-dimethoxy)carbonyl]amino]methylene]amino]ethyl-4-[3-[[Bis[[(1,1-dimethoxy)carbonyl]amino]methylene]amino]-propyl]-2,4-dihydroxy-1[2-[[(1*E*)-2-(4-hydroxyphenyl)-ethenyl]amino]-2-oxoethyl]-5-oxoprolyl-*N*-[(1*E*)-2-(4-hydroxyphenyl)ethenyl]glycinamide (27), (28), and (30). KOH (6 mg, 0.107 mmol) was added to a solution of **26** (28 mg, 0.048 mmol) in 2.5 mL of THF and 2.5 mL of MeOH at 0 °C. The solution was stirred at 0 °C for 20 min, and then 200 μL of 1 M HCl and 5 mL of water were added. The mixture was

extracted with four portions of EtOAc. The combined extracts were dried over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was purified on silica gel (20:1 CH_2Cl_2/MeOH) to give 5 mg (19%) of **28**, followed by 15 mg (58%) of **27**. A trace (~5%) of **30** was seen in the crude ^{1}H NMR spectrum of the crude reaction product. Fractions containing **30** collected from several runs were purified on silica gel (20:1 CH_2Cl_2/MeOH) to give pure **30**.

Data for **27**: ^{1}H NMR (CD_3OD) 7.27 (d, 1, J = 14.8), 7.21 (d, 1, J = 14.8), 7.13 (d, 2, J = 8.8), 7.13 (d, 2, J = 8.8), 6.69 (d, 2, J = 8.8), 6.68 (d, 2, J = 8.8), 6.16 (d, 1, J = 14.8), 6.13 (d, 1, J = 14.8), 4.24 (d, 1, J = 16.8), 4.09 (d, 1, J = 16.8), 3.95 (d, 1, J = 16.8), 3.92 (d, 1, J = 16.8), 3.30−3.53 (m, 4), 2.72 (dd, 1, J = 5.6, 8.4), J = 1.97 (m, 2), 1.72−1.90 (m, 3), 1.51 (m, 1), 1.50 (s, 9), 1.49 (s, 9), 1.47 (s, 9), 1.45 (s, 9); ^{13}C NMR (CD_3OD) 178.1, 173.8, 168.4, 168.2, 164.7, 164.6, 157.9, 157.8 (3 C) 154.3 (2 C), 129.2, 129.0, 128.1 (2 C), 127.9 (2 C), 121.5, 121.1, 116.7 (2 C), 116.6 (2 C), 116.1, 115.2, 91.7, 84.6, 84.5, 80.6, 80.5, 77.1, 46.5, 45.4, 43.9, 42.0, 40.0, 34.7, 28.78 (3 C), 28.76 (3 C), 28.44 (3 C), 28.40 (3 C), 25.0, 24.3; ^{1}H NMR (DMSO-d_6) 11.49 (s, 1, OH), 11.47 (s, 1, OH), 10.19 (d, 1, J = 10.0, NH), 10.10 (d, 1, J = 10.0, NH), 9.39 (s, br, 1, NH), 9.35 (s, br, 1, NH), 8.65 (t, 1, J = 6.3, NH), 8.27 (br, 1, NH), 8.26 (br, 1, NH), 7.15 (dd, 1, J = 10.0, 15.1), 7.14 (d, 2, J = 8.3), 7.13 (d, 2, J = 8.3), 7.12 (dd, 1, J = 10.0, 15.1), 6.68 (d, 2, J = 8.3), 6.66 (d, 1, J = 8.3), 6.63 (s, 1, OH), 6.10 (d, 1, J = 15.1), 6.06 (d, 1, J = 15.1), 5.41 (s, 1, OH), 4.09 (d, 1, J = 17.1), 3.93 (dd, 1, J = 6.3, 16.6), 3.82 (d, 1, J = 17.1), 3.77 (dd, 1, J = 6.3, 16.6), 3.28 (m, 4), 2.53 (1, obscured by DMSO-d_6), 1.78 (m, 3), 1.62 (m, 3), 1.45 (s, 18), 1.39 (s, 9), 1.38 (s, 9), 1.38(m, 1); IR (KBr) 3334, 1719, 1654, 1618.

Data for **28**: ^{1}H NMR (CD_3OD) 7.29 (d, 1, J = 14.8), 7.24 (d, 1, J = 14.8), 7.14 (d, 2, J = 8.8), 7.13 (d, 2, J = 8.8), 6.70 (d, 2, J = 8.8), 6.68 (d, 2, J = 8.8), 6.18 (d, 1, J = 14.8), 6.15 (d, 1, J = 14.8), 4.13 (d, 1, J = 16.0), 4.05 (s, 2), 3.84 (d, 1, J = 16.0), 3.68 (m, 1), 3.56 (m, 1), 3.41 (t, 2, J = 7.2), 2.49 (dd, 1, J = 4.0, 10.0), 1.84−1.95 (m, 2), 1.67−1.82 (m, 3), 1.59−1.67 (m, 1), 1.52 (s, 9), 1.51 (s, 9), 1.47 (s, 9), 1.46 (s, 9); ^{13}C NMR (CD_3OD) 176.5, 173.8, 167.8, 167.5, 164.7, 164.6, 158.0, 157.81, 157.75, 157.7, 154.3, 154.2, 129.2, 129.1, 128.0 (4 C), 121.4, 121.3, 116.7 (4 C), 115.6, 115.3, 92.9, 84.6, 84.5, 80.7, 80.5, 78.4, 48−50 (obscured by CD_3OD), 43.6, 43.3, 42.0, 40.5, 32.3, 28.77 (3 C), 28.76 (3 C), 28.44 (3 C), 28.42 (3 C), 25.3, 25.1; ^{1}H NMR (DMSO-d_6) 11.53 (s, 1, OH), 11.51 (s, 1, OH), 10.12 (d, 1, J = 9.8, NH), 10.04 (d, 1, J = 9.8), 9.36 (s, 1, NH), 9.35 (s, 1, NH), 8.93 (t, 1, J = 5.6, NH), 8.47 (t, 1, J = 5.6, NH), 8.27 (t, 1, J = 5.6), 7.45 (s, 1, OH), 7.21 (dd, 2, J = 9.8, 14.6), 7.14 (d, 2, J = 8.4), 7.13 (d, 2, J = 8.4), 6.67 (d, 4, J = 8.4), 6.094 (d, 1, J = 14.6), 6.087 (d, 1, J = 14.8), 5.34 (s, 1, OH), 4.01 (d, 1, J = 16.6), 3.96 (dd, 1, J = 5.6, 16.4), 3.83 (dd, 1, J = 5.6, 16.4), 3.56 (d, 1, J = 16.6), 3.44 (m, 2), 3.30 (m, 2), 2.42 (dd, 1, J = 3.2, 9.6), 1.48−1.72 (m, 6), 1.47 (s, 18), 1.39 (s, 18); IR (KBr) 3428, 1654.

Data for **30**: ^{1}H NMR (CD_3OD) 7.26 (d, 1, J = 14.8), 7.23 (d, 1, J = 14.8), 7.17 (d, 1, J = 8.8), 7.13 (d, 1, J = 8.8), 6.71 (d, 2, J = 8.8), 6.68 (d, 2, J = 8.8), 6.20 (d, 1, J = 14.8), 6.13 (d, 1, J = 14.8), 4.45 (d, 1, J = 17.2), 4.09 (d, 1, J = 16.8), 3.94 (d, 1, J = 16.8), 3.92 (d, 1, J = 17.2), 3.47 (t, 2, J = 6.8), 3.40 (m, 2), 2.69 (dd, 1, J = 7.2, 7.6), 1.72−2.03 (m, 6), 1.51 (s, 9), 1.51 (s, 9), 1.47 (s, 9), 1.44 (s, 9); ^{13}C NMR (CD_3OD) 178.8, 173.5, 169.0, 168.2, 164.7, 164.7, 158.0, 157.8, 157.8, 157.8, 154.3, 153.7, 129.2, 128.9, 128.2 (2 C), 127.9 (2 C), 121.4, 121.1, 116.7 (4 C), 116.4, 115.2, 90.1, 84.5, 84.5, 80.6, 80.4, 77.5, 51.9, 48−50 (obscured by CD_3OD), 45.4, 43.8, 42.1, 39.9, 33.1, 28.8 (6 C), 28.4 (6 C), 24.5; ^{1}H NMR (DMSO-d_6) 11.50 (s, 1, OH), 11.49 (s, 1, OH), 10.54 (d, 1, J = 9.6, NH), 10.15 (d, 1, J = 9.6, NH), 9.46 (s, 1, NH), 9.39 (s, 1, NH), 8.70 (t, 1, J = 5.6), 8.32 (br, 2, NH), 7.17 (d, 2, J = 8.6), 7.16 (dd, 1, J = 9.6, 14.8), 7.14 (dd, 1, J = 9.6, 14.8), 7.12 (d, 2, J = 8.6), 6.86 (s, 1, OH), 6.69 (d, 2, J = 8.6), 6.67 (d, 2, J = 8.6), 6.15 (d, 1, J = 14.8), 6.08 (d, 1, J = 14.8), 5.58 (s, 1, OH), 4.31 (d, 1, J = 17.1), 3.93 (dd, 1, J = 5.6, 16.6), 3.82 (d, 1, J = 17.1), 3.79 (dd, 1, J = 5.6, 16.6), 3.31 (m, 4), 2.54 (1, obscured by DMSO-d_6), 1.52−1.90 (m, 6), 1.46 (s, 9), 1.45 (s, 9), 1.39 (s, 9), 1.37 (s, 9).

(2α,3α,4α)-3-[2-[(Aminoiminomethyl)amino]-ethyl-4-[3-[(aminoiminomethyl)amino]-propyl]-2,4-dihydroxy-1[2-[[(1*E*)-2-(4-hydroxyphenyl)-ethenyl]amino]-2-oxoethyl]-5-oxoprolyl-*N*-[(1*E*)-2-(4-hydroxyphenyl)ethenyl]glycinamide (Anchinopeptolide D, 4). A solution of **27** (11 mg, 9.34 μmol) in 2 mL of CH_2Cl_2 and 2 mL of trifluoroacetic acid was stirred at 25 °C for 1 h. The mixture was evaporated to dryness to give 11.5 mg (91%) of the TFA salt of **4**: ^{1}H NMR (CD_3OD) 7.25 (d, 1, J = 14.8), 7.22 (d, 1, J = 14.8), 7.15 (d, 4, J = 8.8), 6.71 (d, 4, J = 8.8), 6.18 (d, 1, J = 14.8), 6.17 (d, 1, J = 14.8), 4.22 (d, 1, J = 16.4), 4.13 (d, 1, J = 16.4), 3.92 (d, 1, J = 16.4), 3.90 (d, 1, J = 16.4), 3.46 (m, 1), 3.25 (m, 1), 3.20 (t, 2, J = 6.8), 2.70 (dd, 1, J = 4.2, 9.8), 1.98−2.13 (m, 1), 1.74−1.93 (m, 4), 1.45−1.61 (m, 1); ^{13}C NMR (CD_3OD) 178.0, 173.7, 168.4, 168.1, 158.8, 158.8, 158.0, 157.9, 129.0, 128.9, 128.03 (2 C), 127.96 (2 C), 121.1, 120.9, 116.7 (4 C), 116.2, 115.7, 91.3, 76.8, 46.8, 45.2, 43.5, 42.6, 40.9, 34.7, 24.7, 24.1; ^{1}H NMR (DMSO-d_6) 10.33 (d, 1, J = 10, NH), 10.20 (d, 1, J = 10, NH), 9.47 (s, 1, OH), 9.45 (s, 1, OH), 8.76 (t, 1, J = 6.0, NH), 7.85 (br, 1, NH), 7.70 (br, 1, NH), 6.70−7.50 (br, 8, guanidine NH), 7.17 (dd, 1, J = 10.0, 14.8), 7.15 (d, 4, J = 8.8), 7.14 (dd, 1, J = 10.0, 14.8), 6.97 (s, 1, OH), 6.69 (d, 2, J = 8.8), 6.68 (d, 2, J = 8.8), 6.17 (d, 1, J = 14.8), 6.09 (d, 1, J = 14.8), 5.59 (s, 1, OH), 4.05 (d, 1, J = 16.4), 3.88 (br, 2), 3.75 (d, 1, J = 16.4), 3.23 (m, 1), 3.10 (m, 3), 2.58 (dd, 1, J = 5.2, 10), 1.82 (m, 1), 1.29−1.78 (m, 5); ^{13}C NMR (DMSO-d_6) 175.3, 171.0, 166.5, 166.1, 156.9, 156.8, 156.4, 156.2, 127.0, 126.8, 126.6 (2 C), 126.5 (2 C), 120.4, 120.1, 115.57 (2 C), 115.56 (2 C), 113.3, 112.5, 89.2, 74.4, 44.7, 43.9, 42.3, 38−42 (2 carbons, obscured by DMSO-d_6), 34.0, 23.4, 23.1; IR (KBr) 3375, 1677, 1650. The spectral data are identical to those reported for the natural product.[2,18]

(2α,3β,4β)-3-[2-[(Aminoiminomethyl)amino]ethyl-4-[3-[(aminoiminomethyl)amino]propyl]-2,4-dihydroxy-1[2-[[(1*E*)-2-(4-hydroxyphenyl)ethenyl]-amino]-2-oxoethyl]-5-oxoprolyl-*N*-[(1*E*)-2-(4-hydroxyphenyl)-ethenyl]glycinamide (*epi*-Anchinopeptolide D, 29). A solution of **28** (12 mg, 10.2 μmol) in 3 mL of CH_2Cl_2 and 3 mL of trifluoroacetic acid was stirred at 25 °C for 1 h. The mixture was evaporated to dryness to give 9.5 mg (94%) of the TFA salt of **29**: ^{1}H NMR (CD_3OD) 7.27 (d, 1, J = 14.8), 7.23 (d, 1, J = 14.8), 7.16 (d, 2, J = 8.8), 7.14 (d, 2, J = 8.8), 6.71 (d, 2, J = 8.8), 6.70 (d, 2, J = 8.8), 6.193 (d, 1, J = 14.8), 6.187 (d, 1, J = 14.8), 4.08 (d, 1, J = 16.4), 4.08 (d, 1, J = 16.4), 3.96 (d, 1, J = 16.4), 3.85 (d, 1, J = 16.4), 3.50 (m, 1), 3.38 (m, 1), 3.23 (t, 2, J = 6.4), 2.40 (dd, 1, J = 4.8, 9.2), 1.60−1.94 (m, 6); ^{13}C NMR (CD_3OD) 176.5, 173.1, 168.0, 167.7, 158.8, 158.8, 157.9, 157.9, 129.1, 128.9, 128.0 (4 C), 121.2, 121.1, 116.7 (4 C), 116.0, 115.6, 92.6, 78.1, 51.1, 43.4, 43.3, 42.6, 41.2, 32.2, 24.9, 24.8; ^{1}H NMR (DMSO-d_6) 10.22 (d, 1, J = 10, NH), 10.17 (d, 1, J = 10, NH), 9.44 (s, 1, OH), 9.43 (s, 1, OH), 8.96 (t, 1, J = 6.0, NH), 7.61 (br, 1, NH), 7.49 (br, 1, NH), 7.44 (s, 1, NH), 6.90−7.40 (br, 8 H, guanidine NH), 7.19 (dd, 1, J = 10.4, 14.8), 7.15 (d, 4, J = 8.4), 7.13 (dd, 1, J = 10.0, 14.8), 6.69 (d, 4, J = 8.4), 6.12 (d, 1, J = 14.8), 6.10 (d, 1, J = 14.8), 5.48 (s, 1, OH), 4.01 (d, 1, J = 16.0), 3.90 (t, 2, J = 6.0), 3.64 (d, 1, J = 16.0), 3.27 (m, 1), 3.12 (m, 3), 2.34 (dd, 1, J = 4.4, 8.8), 1.30−1.75 (m, 6); ^{13}C NMR (DMSO-d_6) 173.6, 171.3, 165.7, 165.4, 156.8, 156.7, 156.3, 156.2, 126.9, 126.9, 126.5 (4 C), 120.5, 120.3, 115.6 (4 C), 112.8, 112.5, 90.8, 76.0, 48.2, 42.3, 42.0, 40.8, 31.2, 27.6, 23.6, 23.3.

[3-[(1*R*,2*S*,2a*S*,5*S*,7a*S*,8*R*,9*S*,14a*R*)-8-[2-[(Aminoiminomethyl)amino]ethyl]hexadecahydro-7a.9-dihydroxy-1,2-bis(4-hydroxyphenyl)4,7,10,13-tetraoxocyclobut[*h*]-pyrrolo[1,2-*a*][1,4,7,10]tetraazacyclododeciny-9-yl]-propyl]-guanidine (Cycloanchinopeptolide D, 31). A solution of the TFA salt of anchinopeptolide D (**4**) (10.5 mg, 11.4 μmol) in 2 mL of D_2O in an NMR tube was degassed and purged with N_2 several times and then irradiated at 28 °C with fifteen 350 nm light bulbs for 4.75 h. The solution was evaporated to dryness under reduced pressure, and the residue was purified by flash chromatography on C_{18}-coated silica gel (92.5:7.5 H_2O/MeOH) to give 5 mg (48%) of **31** which decomposed slowly in CD_3OD at 25 °C, probably by equilibration at the hemiaminal center: ^{1}H NMR (CD_3OD) 6.79 (d, 2, J = 8.8), 6.72 (d, 2, J =

8.8), 6.55 (d, 2, J = 8.8), 6.52 (d, 2, J = 8.8), 4.83 (dd, 1, J = 6.7, 7.3), 4.56 (dd, 1, J = 7.3, 7.9), 4.38 (d, 1, J = 13.4), 4.24 (d, 1, J = 17.1), 4.11 (dd, 1, J = 7.9, 10.3), 4.00 (dd, 1, J = 6.7, 10.3), 3.95 (d, 1, J = 17.0), 3.42 (d, 1, J = 13.4), 3.21 (t, 2, J = 7.2), 3.10 (dd, 1, J = 5.2, 9.2), 3.16 (m, 2), 1.78−1.96 (m, 5), 1.62 (m, 1); ^{13}C NMR (CD_3OD) (from HSQC and HMBC experiments) 178.3, 173.8, 172.0, 170.4, 158.8, 158.8, 157.0, 157.0, 130.7, 130.4, 130.0 (4 C), 116.0 (2 C), 115.9 (2 C), 90.6, 76.5, 53.7, 52.4, 47.9, 47.9, 45.7, 43.9, 42.3 (2 C), 40.7, 34.6, 24.5, 24.4.

(*E*)-L-[*N*-Boc-α-glutamyl]-*N*-[2-(4-hydroxyphenyl)ethenyl]glycinamide *tert*-Butyl Ester (33). A mixture of BOC-Glu(OtBu)OH (**32**) (25.3 mg, 83 μmol), DCC (19.0 mg, 92 μmol), and **21** (19.5 mg, 83 μmol) in 1.5 mL of CH_2Cl_2 was stirred at 25 °C for 1 h. The solution was filtered to remove dicyclohexylurea, and the filtrate was evaporated to dryness. MeOH (2 mL), EtOAc (2 mL), and saturated Na_2CO_3 aqueous solution (0.5 mL) were added to the residue, and the mixture was stirred at room temperature for 40 min. 1 N HCl was added until the pH was ∼2. The mixture was concentrated under reduced pressure. The residue was extracted with four portions of EtOAc. The combined extracts were dried over Na_2SO_4. The solvent was evaporated and the residue was purified on silica gel (1:1 EtOAc/CH_2Cl_2) to give 29 mg (71%) of **33**: ^{1}H NMR (CD_3OD) 7.26 (d, 1, J = 14.8), 7.15 (d, 2, J = 8.8), 6.71 (d, 2, J = 8.8), 6.35 (d, 1, J = 14.8), 4.01 (dd, 1, J = 5.6, 8.4), 3.98 (d, 1, J = 17.2), 3.89 (d, 1, J = 17.2), 2.37 (t, 2, J = 7.2), 2.04 (m, 1), 1.90 (m, 1), 1.48 (s, 9), 1.46 (s, 9); ^{13}C NMR (CD_3OD) 175.5, 174.1, 169.0, 158.5, 157.8, 129.1, 127.9 (2 C), 121.0, 116.7 (2 C), 116.1, 82.0, 81.2, 56.3, 43.5, 32.7, 28.8 (3 C), 28.5 (3 C), 27.9; IR (KBr) 3308, 1691, 1655, 1604; $[\alpha]^{25}_D$ +34.4 (*c* 0.625, MeOH).

(*E*)-L-α-Glutamyl-*N*-[2-(4-hydroxyphenyl)ethenyl]glycinamide (34). Protected tripeptide **33** (11 mg, 24 μmol) was dissolved in 0.5 mL of CH_2Cl_2 and 0.5 mL of TFA. The solution was stirred at room temperature for 1 h. The solution was evaporated to dryness under reduced pressure to give 10 mg (100%) of **34**: ^{1}H NMR (CD_3OD) 7.25 (d, 1, J = 14.8), 7.15 (d, 2, J = 8.6), 6.71 (d, 2, J = 8.6), 6.18 (d, 1, J = 14.8), 4.006 (1, obscured by CH_2 singlet at 4.005), 4.005 (s, 2), 2.56 (t, 2, J = 8.0), 2.16 (m, 2); ^{13}C NMR (CD_3OD) 175.9, 170.5, 168.3, 157.8, 129.1, 127.9 (2 C), 121.2, 116.6 (2 C), 115.5, 53.9, 43.3, 30.2, 27.8; IR (KBr) 3327 (br), 3088 (shoulder), 1672 (br); $[\alpha]^{25}_D$ +40.6 (*c* 0.26, MeOH).

(*E*)-L-Pyroglutamyl-*N*-[2-(4-hydroxyphenyl)ethenyl]-glycinamide (35). A mixture of L-pyroglutamic acid (11 mg, 85 μmol), DCC (19.4 mg, 94 μmol), and **21** (20 mg, 85 μmol) in 1 mL of CH_3CN was stirred at room temperature for 40 min. The solution was filtered to remove dicyclohexylurea, and the filtrate was evaporated to dryness. MeOH (2 mL), EtOAc (2 mL), and saturated Na_2CO_3 aqueous solution (0.5 mL) were added, and the mixture was stirred at room temperature for 40 min. 1 N HCl was added until the pH was ∼2. The mixture was concentrated under reduced pressure, and the residue was extracted with six portions of EtOAc. The combined extracts were dried over Na_2SO_4. The solvent was evaporated, and the residue was purified on silica gel (4:1 $CHCl_3$/MeOH) to give 19 mg (73%) of **35**: ^{1}H NMR (CD_3OD) 7.26 (d, 1, J = 14.6), 7.16 (d, 2, J = 8.6), 6.71 (d, 2, J = 8.6), 6.18 (d, 1, J = 14.6), 4.25 (dd, 1, J = 4.9, 8.6), 3.96 (s, 2), 2.52 (m, 1), 2.49 (m, 1), 2.32 (m, 1), 2.15 (m, 1); ^{13}C NMR (CD_3OD) 181.7, 175.8, 168.7, 157.8, 129.2, 127.9 (2 C), 121.2, 116.7 (2 C), 115.5, 58.4, 43.3, 30.6, 26.8; IR (KBr) 3282, 1654; $[\alpha]^{25}_D$ +2.9 (*c* 0.58, MeOH) [lit.[3] $[\alpha]^{25}_D$ −4.6 (*c* 1.0, MeOH)]. The NMR spectral data for **34** are identical to those reported for the natural product assigned structure **34**.[18,25]

***(E)*-*N*-[2-(4-Hydroxyphenyl)ethenyl]glycinamide (36).** A solution of **19** (189 mg, 0.65 mmol) in 2.5 mL of CH_2Cl_2 and 2.5 mL of trifluoroacetic acid was stirred at 25 °C for 10 min. The solution was evaporated to dryness. The residue was dissolved in EtOAc and saturated Na_2CO_3 aqueous solution was added until the pH was 10. The mixture was extracted with 6 portions of EtOAc. The combined extracts were dried over Na_2SO_4 and concentrated to give 106 mg (85%) of **36**: mp 180−182 °C dec; ^{1}H NMR (CD_3OD) 7.28 (d, 1, J = 14.8), 7.16 (d, 2, J = 8.4), 6.72 (d, 1, J = 8.4), 6.19 (d, 1, J = 14.8), 3.46 (s, 2); ^{13}C NMR (CD_3OD) 170.8, 157.8, 129.1, 127.9 (2 C), 121.1, 116.7 (2 C), 115.5, 44.3; IR (KBr) 3273, 1672, 1649, 1610.

***N,N′*-[(1α,2α,3β,4β)-2,4-(4-Hydroxyphenyl)-1,3-cyclobutanediyl)bis[glycinamide] (37).** A solution of **36** (10 mg, 52.6 μmol) in methanol (2 mL) was poured onto a Pyrex Petri dish and the methanol was evaporated quickly by blowing N_2 onto the solution. A film of solid formed on the dish surface. The film was irradiated with fifteen 350 nm light bulbs for 15 h. The residue on the dish was purified on silica gel (3:1 $CHCl_3$/MeOH, then MeOH) to give 4.0 mg (40%) of **37**: ^{1}H NMR (CD_3OD) 7.12 (d, 4, J = 8.4), 6.77 (d, 4, J = 8.4), 4.98 (dd, 2, J = 6.8, 8.4), 3.78 (dd, 2, J = 6.8, 8.4) 3.08 (d, 2, J = 16.4), 2.95 (d, 2, J = 16.4); ^{13}C NMR (CD_3OD) 174.9, 157.6, 130.5 (2 C), 129.2, 116.3 (2 C), 50.7, 50.2, 44.9; IR (KBr) 3345, 1663.

Acknowledgment. We thank the Institute of General Medicine, National Institutes of Health, for financial support. We thank Ms. Christine Hofstetter for assistance in obtaining NOESY, HSQC, and HMBC data and Dr. Agostino Casapullo, Dipartimento di Scienze Farmaceutiche, Universitá di Salerno, Italy, for copies of the spectra of anchinopeptolide D, cycloanchinopeptolide C, and the tripeptide.

Supporting Information Available: X-ray structural data for **13b** including CIF file; ^{1}H and ^{13}C NMR spectra for compounds **4**, **13a**, **13b**, **27**−**31**, and **33**−**37**. This material is available free of charge via the Internet at http://pubs.acs.org.

JO991454L