Total Synthesis of VM55599. Utilization of an Intramolecular Diels−Alder Cycloaddition of Potential Biogenetic Relevance

Emily M. Stocking,† Juan F. Sanz-Cervera,‡ and Robert M. Williams*,†

Contribution from the Department of Chemistry, Colorado State University, Fort Collins, Colorado 80523

Received October 22, 1999

Abstract: The total synthesis of VM55599, a natural metabolite of *Penicillium* sp. IMI332995, has been achieved via an intramolecular Diels−Alder cycloaddition of a reverse isoprene moiety across an azadiene system. The diastereoselectivity of the intramolecular Diels−Alder cycloaddition has biogenetic implications and is discussed in the context of the biogenetic relationship of VM55599 to the paraherquamides.

Introduction

VM55599 is a minor secondary metabolite of *Penicillium* sp. IMI332995.[1] This substance was co-isolated with several known members of the paraherquamide family including paraherquamide A, VM54158 (paraherquamide G), VM54159 (paraherquamide E), and VM55594 (paraherquamide F) by Everett et al.[1] In addition, this *Penicillium* strain was found to produce several new metabolites in the paraherquamide family including VM55595, VM55596, and VM55597 (Figure 1).[1] The relative stereochemistry of VM55599 was assigned by ^{1}H NMR/NOE data but the absolute stereochemistry remains unknown; the absolute stereostructure depicted below is a prediction based on biogenetic considerations to be discussed below. Of particular interest in this regard is the stereochemical disposition of the methyl group in the β-methylproline ring which was assigned as being *syn* to the bridging isoprene moiety. In all other known members of the paraherquamide family, the methyl group in the β-methylproline ring is disposed *anti* to the bridging isoprene moiety.

VM55599,[1] the paraherquamides,[2] brevianamides,[3] marcfortines,[4] and most recently, the sclerotamides,[5] are indolic secondary metabolites isolated from various fungi and have attracted considerable attention due to their molecular complexity, intriguing biogenesis,[6] and some members, most notably the paraherquamides, display potent antiparasitic activity.[7] These

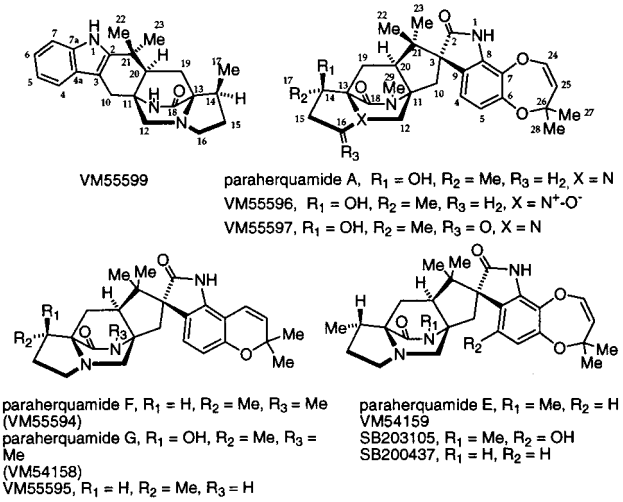


Figure 1.

alkaloids share the unusual bicyclo[2.2.2] ring system that has been proposed to arise via the [4 + 2] cycloaddition of the isoprene moiety across the α-carbons of the amino acid units.[6c,d,8,9] Previous work on the biosynthesis of these substances invoked a facial divergence in the Diels−Alder cyclization which sets the relative *syn*-/*anti*-stereochemical relationship at this stereogenic center.[10,11] Specifically, the cyclization of the isoprenyl olefin across the azadiene ring system can proceed via four distinct diasteromeric transition structures **a**, **b**, **c**, or **d** (Figure 2), resulting in the four corresponding cycloadducts **A**, **B**, **C**, or **D**. Cycloadduct **B** corresponds to VM55599, and cycloadduct **A** is the putative structure leading to paraherqua-

† Department of Chemistry, Colorado State University.

‡ Departamento de Química Orgánica, Universidad de Valencia, E−46100 Burjassot, Spain.

(1) Blanchflower, S. E.; Banks, R. M.; Everett, J. R.; Reading, C. *J. Antibiot.* **1993**, *46*, 1355.

(2) (a) Yamazaki, M.; Okuyama, E.; Kobayashi, M.; Inoue, H. *Tetrahedron Lett.* **1981**, *22*, 135. (b) Ondeyka, J. G.; Goegelman, R. T.; Schaeffer, J. M.; Kelemen, L.; Zitano, L. *J. Antibiot.* **1990**, *43*, 1375. (c) Liesch, J. M.; Wichmann, C. F. *J. Antibiot.* **1990**, *43*, 1380. (d) S. E.; Banks, R. M.; Everett, J. R.; Manger, B. R.; Reading, C. *J. Antibiot.* **1991**, *44*, 492.

(3) (a) Birch, A. J.; Wright, J. J. *J. Chem. Soc., Chem. Commun.* **1969**, 644. (b) Birch, A. J.; Wright, J. J. *Tetrahedron* **1970**, *26*, 2329. (c) Birch, A. J.; Russell, R. A. *Tetrahedron* **1972**, *28*, 2999. (d) Bird, B. A.; Remaley, A. T.; Campbell, I. M. *Appl. Environ. Microbiol.* **1981**, *42*, 521. (e) Bird, B. A.; Campbell, I. M. *Appl. Environ. Microbiol.* **1982**, *43*, 345. (f) Robbers, J. E.; Straus, J. W. *Lloydia* **1975**, *38*, 355. (g) Paterson, R. R. M.; Hawksworth, D. L. *Trans. Br. Mycol. Soc.* **1985**, *85*, 95. (h) Wilson, B. J.; Yang, D. T. C.; Harris, T. M. *Appl. Microbiol.* **1973**, 26, 633. (I) Coetzer, J. *Acta Crystallogr.* **1974**, *B30*, 2254.

(4) (a) Polonsky, J.; Merrien, M.-A.; Prange, T.; Pascard, C. *J. Chem. Soc., Chem. Commun.* **1980**, 601. (b) Prange, T.; Buillion, M.-A.; Vuilhorgne, M.; Pascard, C.; Polonsky, J. *Tetrahedron Lett.* **1980**, 22, 1977.

(5) Whyte, A. C.; Gloer, J. B. *J. Nat. Prod.* **1996**, *59*, 1093.

(6) (a) Baldas, J.; Birch, A. J.; Russell, R. A. *J. Chem. Soc. Perkin Trans 1* **1974**, 50. (b) Birch, A. J. *Agr. Food Chem.* **1971**, *19*, 1088. (c) Sanz-Cervera, J. F.; Glinka, T.; Williams, R. M. *J. Am. Chem. Soc.* **1993**, *115*, 347. (d) Sanz-Cervera, J. F.; Glinka, T.; Williams, R. M. *Tetrahedron* **1993**, *49*, 8471. (e) Kuo, M. S.; Wiley: V. H.; Cialdella, J. I.; Yurek, D. A.; Whaley, H. A.; Marshall, V. P. *J. Antibiot.* **1996**, *49*, 1006.

(7) (a) Shoop, W. L.; Egerton, J. R.; Eary, C. H.; Suhayda, D. *J. Parasitol.* **1990**, *76*, 349. (b) Shoop, W. L.; Michael, B. F.; Haines, H. W.; Eary, C. H. *Vet. Parasitol.* **1992**, *43*, 259. (c) Shoop, W. L.; Haines, H. W.; Eary, C.H.; Michael, B. F. *Am. J. Vet. Res.* **1992**, *53*, 2032. (d) Ostlind, D. A.; Mickle, W. G.; Ewanciw, D. V.; Andriuli, F. J.; Campbell, W. C.; Hernandez, S.; Mochales, S.; Munguira, E. *Res. Vet. Sci.* **1990**, *48*, 260. (e) Schaeffer, J. M.; Blizzard, T. A.; Ondeyka, J.; Goegelman, R.; Sinclair, P. J.; Mrozik, H. *Biochem. Pharmacol.* **1992**, *43*, 679.

(8) Porter, A. E. A.; Sammes, P. G. *J. Chem. Soc., Chem. Commun.* **1970**, 1103.

10.1021/ja993791h CCC: $19.00
Published on Web 02/04/2000

mide; cycloadducts **C** and **D** lead to C-20-*epi* metabolites thus far not detected in paraherquamide-producing fungi.

In addition, recent theoretical work on an indoxyl-based Diels−Alder cyclization pathway supported the observed isomer distribution of the brevianamides in *Penicillium brevicompactum* which produces brevianamide A as the major metabolite and brevianamide B as the minor metabolite, both of which possess the *anti*-relationship.[12] As part of a program directed primarily at elucidating the biosynthetic mechanism of formation of the unique bicyclo[2.2.2] ring system, particularly with respect to the question of possible enzymatic catalysis of this reaction, we report here the first total synthesis of VM55599 using an intramolecular Diels−Alder cyclization reaction that may be of biogenetic relevance.[13]

Results and Discussion

The synthesis of VM55599 was accomplished as shown in Scheme 1. The benzophenone imine **1** of glycine ethyl ester was condensed with the dimethylallylated gramine derivative **2**[14] in the presence of tri-*n*-butylphosphine[15] in acetonitrile to furnish the tryptophan derivative **3** in 70% yield. Cleavage of the benzophenone imine with hydroxylamine provided the amino ethyl ester **4** in high yield. Subsequent *t*-BOC protection and basic hydrolysis of the ethyl ester furnished the acid **5** in 78% yield over two steps. Coupling of acid **5** with racemic β-methyl-β-hydroxyproline ethyl ester with BOP reagent[16] provided the desired dipeptide **7** in 70−83% yield. The BOC group was cleaved with TFA, and the resulting amino ethyl ester was cyclized to the corresponding piperazinedione **8** in the presence of 2-hydroxypyridine in refluxing toluene in excellent yield.

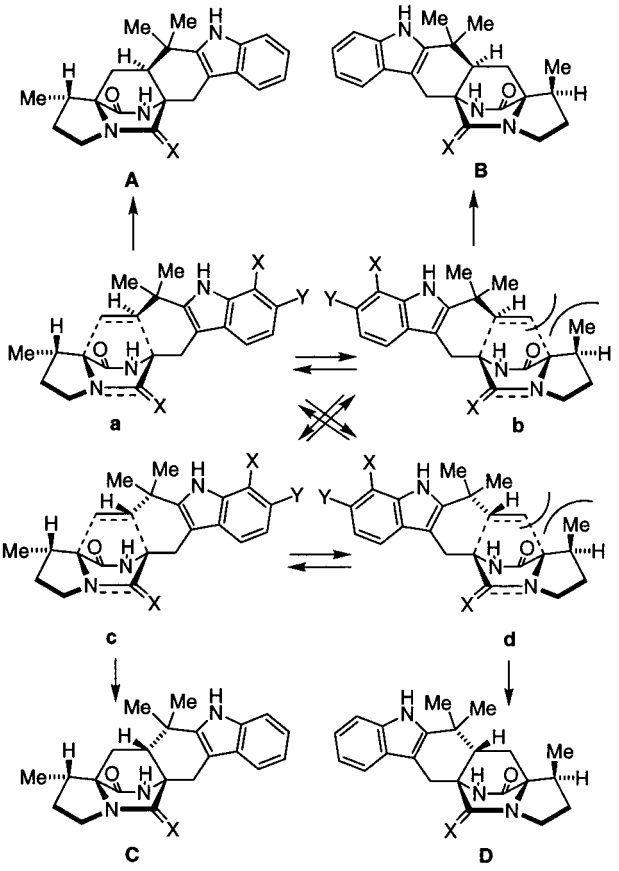
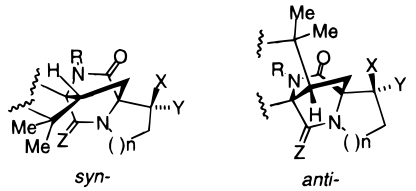


Figure 2.

Treatment of **8** with thionyl chloride in pyridine furnished the unsaturated substance **9** in 75% yield. Subsequent treatment of **9** with trimethyloxonium tetrafluoroborate in dichloromethane provided the azadiene **10** in 72% yield. Treatment of azadiene **10** with KOH in aqueous methanol effected tautomerization to the labile incipient azadiene **11** which spontaneously suffered intramolecular Diels−Alder cycloaddition at room temperature to give a mixture of all four possible racemic cycloadducts **12**−**15** in 78% combined yield in a 3.7:2.6:1.6:1 ratio, respectively.

(9) For reviews on biosynthetic Diels−Alder reactions, see: (a) Tarasow, T. M.; Eaton, B. E. *CMLS Cell Mol. Life Sci.* **1999**, 1463 (b) Laschat, S. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 289. (c) Stipanovic, R. D. *Environ. Sci. Res.* **1992**, *44*, 319. For specific references on Diels−Alder reactions proposed in natural products biosynthesis, see: (d) catharanthin: Scott, A. I. *Acc. Chem. Res.* **1970**, *3*, 151. (e) Ikarugamycin: Ito, S.; Hirata, Y. *Tetrahedron Lett.* **1972**, *25*, 2557. (f) Cytochalasans: Binder, M.; Tamm, C. *Angew. Chem., Int. Ed. Engl.* **1973**, *12*, 370. (g) Ircinianin: Hofheinz, W.; Schonholzer, P. *Helv. Chim. Acta* **1977**, *60*, 1367. (h) lachananthocarpone: Bazan, A. C.; Edwards, J. M., Weiss, U. *Tetrahedron* **1978**, *34*, 3005. (i) Endiandric acids: Bandaranayake, W. M.; Banfield, J. E., Black, D. S. C. *J. Chem. Soc., Chem. Commun.* **1980**, 902. (j) Antibiotic X-14547A: Roush, W. R.; Peseckis, S. M.; Walts, A. E. *J. Org. Chem.* **1984**, *49*, 3429. (k) Mevinolin: Moore, R. N.; Bigam, G.; Chan, J. K.; Hogg, A. M. Nakashima, T. T.; Vederas, J. C. *J. Am. Chem. Soc.* **1985**, *107*, 3694. Witter, D. J.; Vederas, J. C. *J. Org. Chem.* **1996**, *61*, 2613. Kennedy, J.; Auclair, K.; Kendrew, S. G.; Park, C.; Vederas, J. C.; Hutchinson, C. R. *Science* **1999**, *284*, 1368. (l) Bishordeninyl terpenes: Schroeder, D. R.; Stermitz, F. R. *Tetrahedron* **1985**, *41*, 4309. (m) Coumarins: Dreyer, D. L. *Recent Adv. Phytochem.* **1986**, *20*, 317 (n) Betaenone B: Harrison, P. H. M.; Cane, D. E. *Chemtracts: Org. Chem.* **1988**, *1*, 369. (0) Sordaricin: Wu, X.; Kato, N.; Takeshita, H. *Tennen Yuki Kagobutsu Toronkai Koen Yoshishu* **1988**, *30*, 424. (p) Esperamicins: Schreiber, S. L.; Kiessling, L. L. *J. Am. Chem. Soc.* **1988**, *110*, 631. (q) Solanapyrone A: Oikawa, H.; Yokota, T.; Abe, T.; Ichihara, A.; Sakamura, S.; Yoshizawa, Y.; Vederas, J. C. *J. Chem. Soc., Chem. Commun.* **1989**, 1282 and Oikawa, H.; Suzuki, Y.; Katayama, K.; Naya, A.; Chiaki, S.; Ichihara, A. *J. Chem. Soc., Perkin Trans 1* **1999**, 1225 and references therein. (r) Kuwanon J and chalcomoracin: Hano, Y. Nomura, T.; Ueda, S. *Chem. Pharm. Bull.* **1989**, *37*, 554. Hano, Y.; Nomura, T.; Ueda, S. *Heterocycles* **1999**, *51*, 231. (s) Plagiospirolides C, D and E: Spoerle, J.; Becker, H.; Gupta, M. P.; Veith, M.; Huch, V. *Tetrahedron* **1989**, *45*, 5003. (t) Eudesmanolides: Marco, J. A.; Sanz, J. F.; Falco, E. *Tetrahedron* **1990**, *46*, 7941. Manzamines: Baldwin, J. E.; Whitehead, R. C. *Tetrahedron Lett.* **1992**, *33*, 2059. Baldwin, J. E. et al. *Angew. Chem., Int. Ed.* **1998**, *37*, 2661 and references therein. (u) Theonellastreone and conicasterone: Kobayashi, M.; Kawazoe, K., Katori, T.; Kitagawa, I. *Chem. Pharm. Bull.* **1992**, *40*, 1773. (v) Nargenicin: Cane, D. E.; Yang, C.-C. *J. Am. Chem. Soc.* **1984**, *106*, 784. Cane, D. E.; Tan, W.; Ott, W. R. *J. Am. Chem. Soc.* **1993**, *115*, 527. (w) Torreyanic acid: Lee, J. C.; Strobel, G. A.; Lobovsky, E.; Clardy, J. *J. Org. Chem.* **1996**, *61*, 3232. (x) Cochleamycins: Shindo, K.; Sakakibara, M.; Kawai, H.; Seto, H. *J. Antibiot.* **1996**, *49*, 249. (y) Wistarin: Fontana, A.; Fakhr, I.; Mollo, E.; Cimino, G. *Tetrahedron: Asymmetry* **1999**, *10*, 3869. (z) bisorbicillinol: Nicolaou, K. C.; Simonsen, K. B.; Vassilikogiannakis, G.; Baran, P. S.; Vidali, V. P.; Pitsinos, E. N.; Couladouros, E. A. *Angew. Chem., Int. Ed.* **1999**, *38*, 3555 and references therein.

(10) The *syn*/*anti* relationship refers to the relative stereochemistry between the C-20 stereogenic center (VM55599 numbering) and the cyclic amino acid residue (proline, β-methylproline, or pipecolic acid):



(11) Williams, R. M.; Kwast, E.; Coffman, H,; Glinka, T. *J. Am. Chem. Soc.* **1989**, *111*, 3064.

(12) Domingo, L. R.; Sanz-Cervera, J. F.; Williams, R. M.; Picher, M. T.; Marco, J. A. *J. Org. Chem.* **1997**, *62*, 1662.

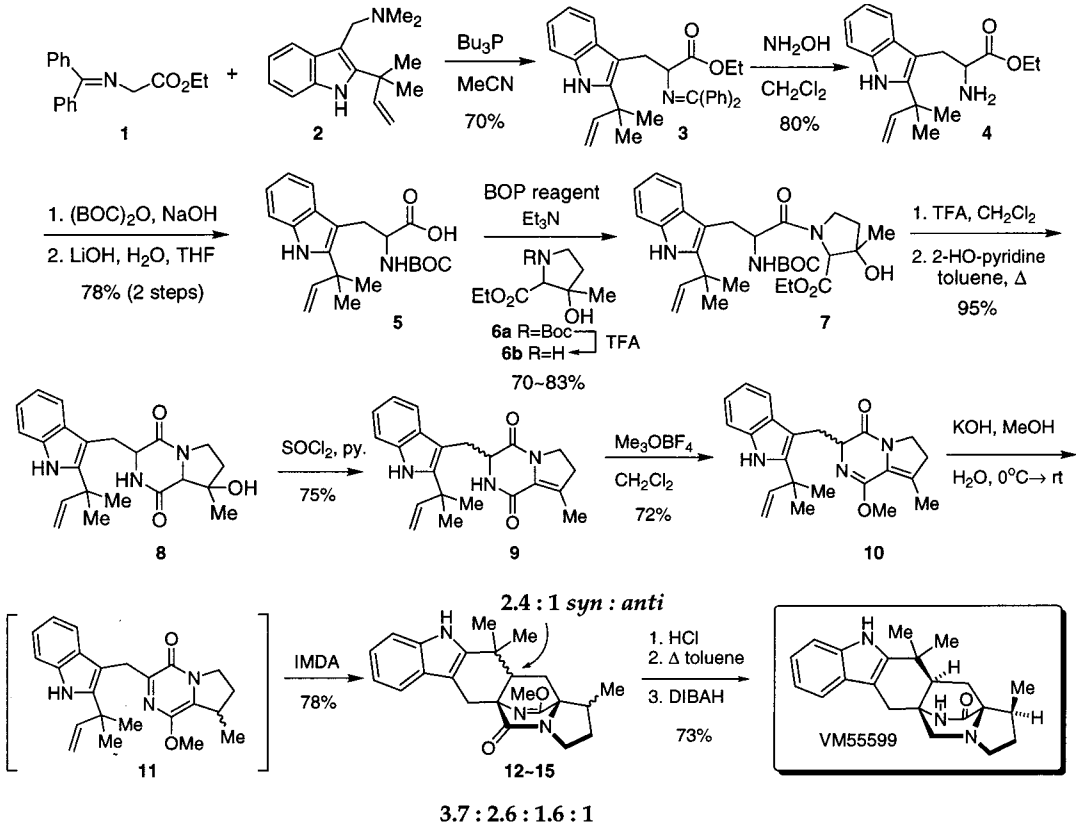
(13) (a) Williams, R. M.; Sanz-Cervera, J. F.; Sancenon, F., Marco, J. A.; Halligan, K. *J. Am. Chem. Soc.* **1998**, *120*, 1090. (b) Williams, R. M.; Sanz-Cervera, J. F.; Sancenon, F., Marco, J. A.; Halligan, K. *Bioorg. Med. Chem.* **1998**, *6*, 1233.

(14) This substance was prepared according to ref 6d.

(15) (a) Kametani, T.; Kanaya, N.; Ihara, M. *J. Chem. Soc., Perkin Trans 1* **1981**, 959. (b) Somei, M.; Karasawa, Y.; Kaneko, C. *Heterocycles* **1981**, *16*, 941.

(16) BOP = (benzotriazol-1-yloxy)tris(dimethylamino)phosphonium hexafluorophosphate (purchased from Aldrich Chemical Co.).

Scheme 1



The Diels−Alder cycloadducts **12**−**15** were separable by PTLC on silica gel, and their relative stereochemistry was assigned by ^{1}H NMR NOE studies.[17] The *syn*-stereochemistry at C-20 for **12** and **13** was assigned based on the NOE between H-20 (using the VM55599 numbering system) and the OMe of the lactim ether. The *anti*-stereochemistry assignment of C-20 for both **14** and **15** was made based on the NOE between H-23 and the OMe. The assignment of stereochemistry at C-14 for **12** and **14** was deduced from the NOE between H-17 and H-19/19′. This NOE was also observed by Everett et al. in the original VM55599 isolation paper.[1] For **13** and **15**, the stereochemical assignment of C-14 was inferred from the NOE between H-14 and H-19/19′.

The structures of all four cycloadducts **12**−**15** depicting their relative stereochemistries are shown in Figure 3. The *syn/anti* relationship[10] at the C-20 stereogenic center was 2.4:1 and is consistent with results reported earlier from this laboratory on a simpler system lacking the methyl group in the proline ring.[13] Of significant interest was the unexpected observation that the major products (**12** and **14**) in each diastereomeric subset displayed the methyl group in the β-methylproline ring *syn* to the bridging isoprene unit (see Figure 3). The diastereoselectivity in this regard was 1.47:1 favoring the methyl group disposed *syn* to the bridging isoprene moiety. Although it is reasonable to expect modest diastereoselectivity for this Diels−Alder cycloaddition, purely on the basis of the slight steric bias expected to be exerted by the methyl group in the proline ring, we anticipated a modest preference for cycloadducts that displayed the methyl group *anti* to the bridging isoprene moiety.

Confirmation of the structure for cycloadduct **12** was secured through conversion into racemic VM55599. Thus, treatment of

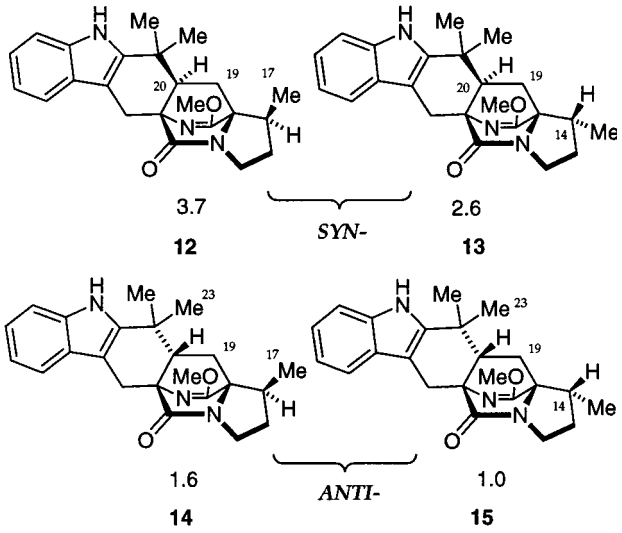


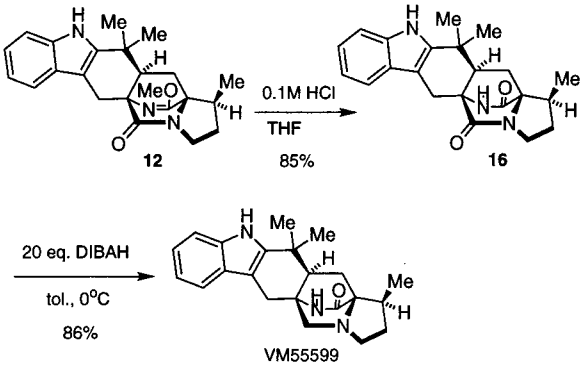
Figure 3.

12 with dilute HCl effected cleavage of the lactim ether to the corresponding secondary amide **16** in 85% yield (Scheme 2). Selective reduction of this substance with excess DIBAH[18] (20 equiv) provided VM55599 in 86% yield whose ^{1}H and ^{13}C NMR spectral characteristics matched those published.[1] The synthetic material was subsequently utilized to guide reisolation of natural VM55599 from cultures of *Penicillium* sp. IMI332995 (obtained from the International Mycological Institute) grown in our laboratory. The synthetic and natural specimens were found to have identical ^{1}H NMR spectra and TLC mobility thereby confirming the assignment (see Supporting Information).

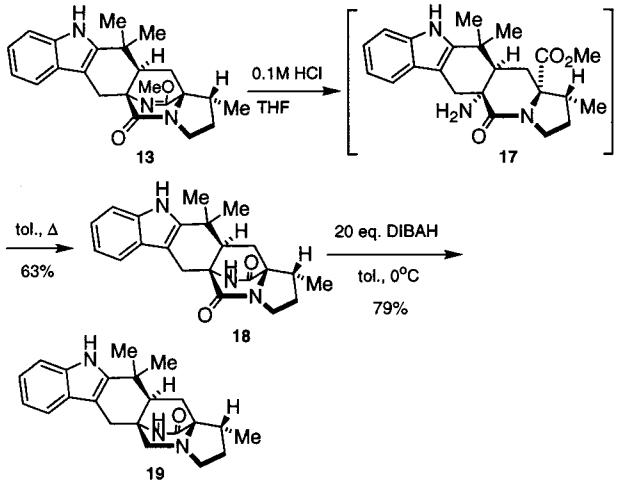
(17) Complete NOE data on cycloadducts **12**−**15** can be found in Supporting Information.

(18) We thank Prof. Tohru Fukuyama for suggesting the use of excess DIBAH for this transformation; see Fukuyama, T.; Liu, G. *Pure Appl. Chem.* **1997**, *69*, 501.

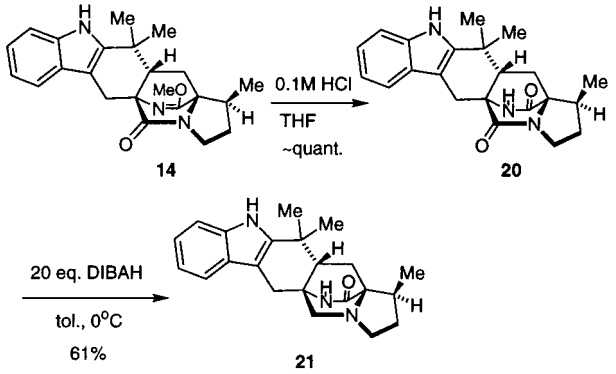
Scheme 2



Scheme 3



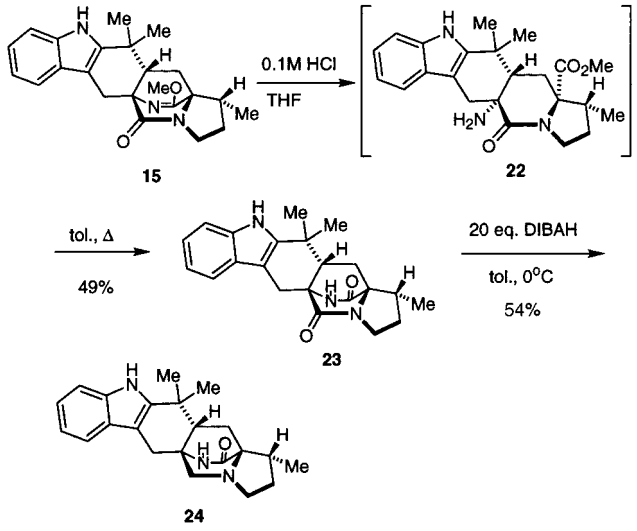
Scheme 4



To further confirm this assignment, the three other cycloadducts **13**−**15** were similarly converted into the corresponding C-14 and/or C-20 epimers of VM55599 (**19**, **21**, and **24**) as shown in Schemes 3−5.[19] It was interesting to observe that, in the case of cycloadducts **13** and **15**, cleavage of the lactim ether with dilute HCl led to the production of the ring-opened amino esters **17** and **22**, respectively. These were readily cyclized to the corresponding bicyclo[2.2.2]-containing secondary amides **18** and **23**, respectively, by simply heating these substances in toluene at reflux temperature overnight. In contrast, the lactim ethers of both cycloadducts **12** and **14** could be cleaved to the corresponding bicyclo[2.2.2]-containing substrates without attendant ring-opening to the corresponding amino esters. It would appear that there is $A^{(1,3)}$-type strain in compounds **13** and **15**

(19) Conditions for the conversion of compounds **13**−**15** to the VM55599 diasteromers **19**, **21**, and **24** were not optimized.

Scheme 5



caused by compression between the methyl group disposed on the β-face of the proline ring and the lactim ether methoxy group that is relieved upon ring-opening to **17** and **22**, respectively. In substrates **12** and **14**, where the methyl group in the proline ring is on the α-face, the opportunity for $A^{(1,3)}$-type strain is obviated by the *anti*-relationship between the lactim ether group and the methyl group. Subsequent DIBAH reduction of the tertiary amides of compounds **18**, **20**, and **23** gave the corresponding diastereomers of VM55599 (**19**, **21**, and **24**, respectively).

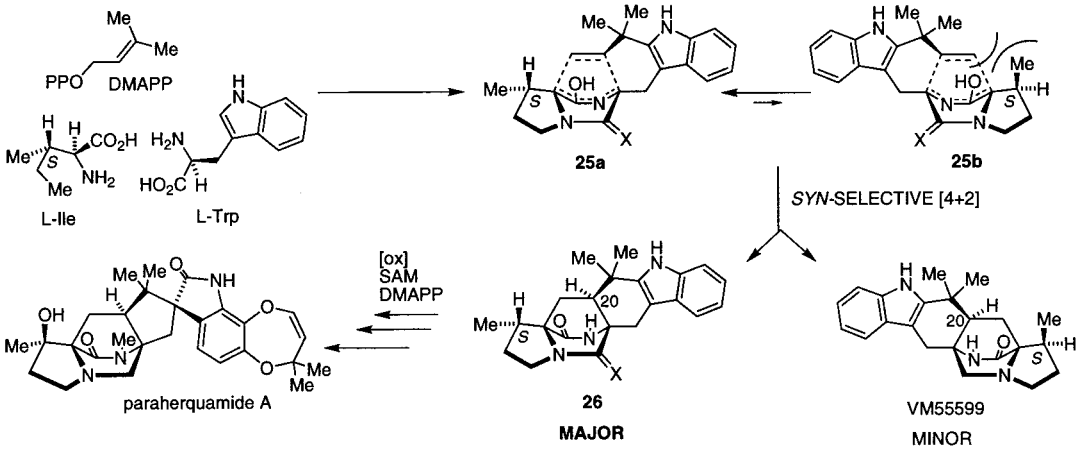
The NMR spectra of the VM55599 diastereomers **19**, **21**, and **24** were fully consistent with the assigned structures, and significantly, all were distinctly different from the spectra for natural VM55599 (see Supporting Information).[1]

A significant implication of these observations concerns the biogenesis and absolute stereochemistry of VM55599. In particular, it should first be noted that natural paraherquamide derivatives containing a non-hydroxylated β-methylproline residue, such as VM55594, VM55595, VM54159, SB203105,[20] and SB200437,[20] all display the methyl group at the β-position of the proline residue *anti* to the bridging isoprene moiety. In stark contrast, VM55599 is the only member of the paraherquamide family thus far isolated that displays the methyl group at the β-position of the proline residue *syn* to the bridging isoprene moiety. We previously demonstrated that the β-methyl-β-hydroxyproline ring of paraherquamide A is biosynthetically derived from L-isoleucine.[21] Enzymatic hydroxylation of the (*S*)-β-methylproline ring in paraherquamide A biosynthesis must therefore occur with net *retention* of stereochemistry leaving the methyl group *anti* to the bridging isoprene moiety. One possible scenario constituting a unified biogenesis of paraherquamide A (and congeners) and VM55599 is depicted in Scheme 6. Since Paraherquamide A and VM55599 both possess the bicyclo[2.2.2] monoketopiperazine ring system and are coproduced by the same fungi, it is tempting to speculate that these substances arise via a related or common [4 + 2] cycloaddition. Thus, if a similar Diels−Alder cyclization, whether it be uncatalyzed or enzyme-catalyzed, is operating in the biosynthetic construction of these metabolites, the isoprene unit must approach the azadiene from the *same* face as the methyl group in the proline ring for VM55599 (**25b**, Scheme

(20) Banks, R. M.; Blanchflower, S. E.; Everett, J. R.; Manger, B. R.; Reading, C. *J. Antibiot.* **1997**, *50*, 840.

(21) Stocking, E.; Sanz-Cervera, J. F.; Williams, R. M.; Unkefer, C. J. *J. Am. Chem. Soc.* **1996**, *118*, 7008.

Scheme 6



6), whereas in paraherquamide A, Diels Alder cyclization must occur from the face *opposite* to the methyl group (**25a**, Scheme 6). In both cases, the diastereofacial selectivity of the Diels−Alder reaction must give the *syn*-relative stereochemistry at C-20 (VM55599 numbering). It is interesting to note that metabolites possessing the *anti*-relative stereochemistry at C-20 have not yet been isolated from paraherquamide-producing fungi. Since VM55599 is a very minor metabolite of *Penicillium* sp. IMI332995,[22] it seems reasonable that a *syn*-selective Diels−Alder reaction gives, via conformer **25a**, cycloadduct **26** as a major product that is then further metabolized to the paraherquamide family. The minor cycloaddition product (via conformer **25b**), after adjustment of the oxidation state at C-12, would furnish VM55599 as a dead-end shunt metabolite with the absolute stereochemistry depicted (predicted).

It is therefore quite interesting that the diastereofacial bias of the Diels−Alder cycloaddition on synthetic azadiene **11** gives a slight preponderance (1.47:1) of cycloaddition from the *same* face as the methyl group in the β-methylproline ring and modest selectivity (2.4:1) favoring the (C-20) *syn*-relative stereochemistry. These results indicate that the *intrinsic* facial bias of this type of Diels−Alder cycloaddition is modest at best and that the biological system may be subject to protein organization of the precyclization conformers.[9]

Conclusion

This study confirms the structural and relative stereochemical assignment made for VM55599[1] and further demonstrates that the core bicyclo[2.2.2] ring system common to this family of alkaloids very likely arises by a biosynthetic intramolecular Diels−Alder cyclization from a preformed dioxopiperazine[23] that subsequently undergoes oxidation to an azadiene species. Finally, the C-20-*epi*-metabolites (with the *anti*-stereochemistry corresponding to the brevianamides) have not yet been detected from paraherquamide-producing fungi, and there have been no reports on the isolation of similarly epimeric metabolites from the brevianamide-producing *Penicillium* sp. Thus, in each biosynthetic system, there appears to be complete facial exclusivity in the construction of the bicyclo[2.2.2] ring nucleus with respect to the relative stereochemistry set at C-20; such is not the case for the laboratory cycloaddition reported here. It is also important to stress that the laboratory cyclization described here occurs spontaneously at room temperature in water, which indicates that the fundamental thermodynamics of this cyclization are amenable to cytosolic constraints. Thus, "catalysis" of this type of cycloaddition may not be required biosynthetically, but the predisposition of the precyclization conformers (ie., **25a**/**25b**) leading to the observed paraherquamide stereoisomers may be a manifestation of incidental protein organization.[9,24] Uncertainties as to the oxidation state of the putative azadiene moiety in the biosynthetic system still exist and are the subject of ongoing investigations in these laboratories.

Experimental Section

***N*-(Diphenylmethylene)-2-(1,1-dimethyl-2-propenyl)-D,L-tryptophan Ethyl Ester (3).** *N*-(Diphenylmethylene)glycine ethyl ester (**1**) (3.2 g, 12.0 mmol) and the gramine derivative **2**[14] (3.2 g, 13.2 mmol) were stirred in acetonitrile (110 mL) under argon until the solids dissolved. Tri-*n*-butylphosphine, (1.5 mL, 6 mmol) was added, and the mixture was brought to reflux temperature for 8 h. After being cooled to room temperature, the solvent was concentrated under reduced pressure, and the crude product was purified by flash silica gel column chromatography (15% EtOAc/hex) to yield 3.78 g (70%) of **3** as sticky yellow foam. ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.29 (3H, t, J = 7.0 Hz), 1.38 (3H, s), 1.41 (3H, s), 3.59 (2H, dd, J = 1.1, 7.0 Hz), 4.23 (2H, m), 4.52 (1H, dd, J = 6.2, 7.3 Hz), 5.04 (1H, dd, J = 1.1, 10.6 Hz), 5.09 (1 H, dd, J = 1.3, 17.2 Hz), 5.92 (1H dd, J = 10.3, 17.2 Hz), 6.39 (2H, bs), 6.87 (1H, ddd, J = 1, 8.3, 8.3 Hz), 7.06 (1H, ddd, J = 1.1, 7.5, 7.5 Hz), 7.09 (2H, dd, J = 1.1, 7.5, 7.5 Hz), 7.30 (5 H, m), 7.45 (1H, d, J = 8.0 Hz), 7.60 (2H, m), 7.86 (1H, bs). ^{13}C NMR (75 MHz, $CDCl_3$): δ 14.1, 27.50, 27.52, 28.7, 38.9, 60.8, 66.6, 107.2, 109.7, 111.5, 118.8, 119.3, 121.1, 127.5, 127.6, 127.7, 128.7, 129.9, 130.0, 133.8, 135.9, 139.2, 139.8, 146.0, 169.7, 172.4. IR (NaCl neat): 3405, 3057, 2972, 1731, 1621, 1597, 1575, 1489, 1462, 1446, 1286, 1245, 1185, 1069, 1029, 917, 781, 742, 697 cm^{-1}. HRMS (FAB+): Calcd for $C_{31}H_{33}N_2O_2$: 465.2542. Found 465.2541 (M + H).

2-(1,1-Dimethyl-2-propenyl)-D,L-tryptophan Ethyl Ester (4). Compound **3** (1.92 g, 4.27 mmol) was stirred with NH_2OH·HCl (2.25 g, 32.45 mmol) and anhydrous Na_2CO_3 (3.21 g, 30.31 mmol) in CH_2Cl_2 (17 mL) at room temperature under argon for 24 h. The solution was acidified to pH 3 with 10% $KHSO_4$ (aq), and the organic layer was separated from the aqueous phase. The aqueous layer was extracted three more times with EtOAc before it was made basic with 10% Na_2CO_3 (aq) and extracted three times with EtOAc. The combined organic layers from the basic extract were dried over anhydrous Na_2SO_4, and the solvent was evaporated under reduced pressure to give 1.03 g (80%) of **4** as a colorless oil. ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.19 (3H, t, J = 7 Hz), 1.52 (2H, bs), 1.58 (6H, s), 3.08 (1H, dd, J = 14.3, 9.5 Hz),

(22) Our own work with *Penicillium* sp. IMI 332995 provided ca. 0.5 mg of VM55599 and ~250 mg of paraherquamide A (~1:500 ratio) from 12 L of surface culture media.

(23) For relevant work, see (a) Dunkerton, L. V.; Chen, H.; McKillican, B. P. *Tetrahedron Lett.* **1988**, *29*, 2539. (b) Fabre, J. L.; Farge, D.; James, C.; Lave, D. *Tetrahedron Lett.* **1985**, *26*, 5447.

(24) For an analogous system, see Oikawa, H.; Katayama, K.; Suzuki, Y.; Ichihara, A. *J. Chem. Soc., Chem. Commun.* **1995**, 1321.

3.35 (1H, dd, $J = 14.7, 5.1$ Hz), 3.86 (1H, dd, $J = 9.5, 5.1$ Hz), 4.13 (2H, m), 5.18 (1 H, dd, $J = 0.7, 10.6$ Hz), 5.20 (1 H, dd, $J = 0.7, 17.2$ Hz), 6.16 (1H, dd, $J = 10.6, 17.2$ Hz), 7.08 (1H, t, $J = 7$ Hz), 7.14 (1H, t, $J = 7$ Hz), 7.29 (1H, d, $J = 7$ Hz), 7.58 (1H, d, $J = 7$ Hz), 7.98 (1H, bs). ^{13}C NMR (75 MHz, $CDCl_3$): δ 14.0, 27.8, 27.9, 31.2, 39.1, 55.9, 60.8, 107.0, 110.3, 112.1, 118.6, 119.3, 112.5, 129.8, 134.1, 140.4, 146.0, 175.5. IR (NaCl neat) 3399, 3243, 3081, 3056, 2973, 1733, 1638, 1617, 1580, 1462, 1300, 1282, 1195, 1105, 1029, 917, 859, 743 cm $^{-1}$. HRMS (FAB+): Calcd for $C_{18}H_{25}N_2O_2$: 301.191603. Found 301.191898 (M + H).

***N*-[(1,1-Dimethylethoxy)carbonyl]-2-(1,1-dimethyl-2-propenyl)-D,L-tryptophan Ethyl Ester.** Compound **4** (1.57 g, 5.23 mmol) was stirred with 1 equiv of 0.5 M NaOH and di-*tert*-butyl pyrocarbonate (1.25 g, 5.75 mmol) in dioxane (5.23 mL) at room temperature for 3 h. The dioxane was removed under reduced pressure, and the solution was brought to pH = 2 with the addition of aqueous 10% $KHSO_4$. The aqueous layer was extracted three times with EtOAc and dried over anhydrous Na_2SO_4. After removing the solvent under reduced pressure, the product was purified by flash silica gel column chromatography using 30% EtOAc/hexane to yield 1.843 g (88%) of the product as an oil. ^{1}H NMR (300 MHz, DMSO-d_6, 120 °C): δ 1.04 (3H, t, $J = 7.0$ Hz), 1.31 (9H, s), 1.55 (6H, s), 3.12 (1H, dd, $J = 7.7, 14.3$ Hz), 3.30 (1H, dd, $J = 6.8, 14.8$ Hz), 3.96 (2H, m), 4.30 (1H, dd, $J = 7.7, 15.6$ Hz), 5.07 (1H, dd, $J = 0, 10.3$ Hz), 5.10 (1H, dd, $J = 0, 17.6$ Hz), 6.22 (1H, dd, $J = 10.7, 17.6$ Hz), 6.26 (1H, bs), 6.92 (1H, t, $J = 7.3$ Hz), 7.00 (1H, t, $J = 7.0$ Hz), 7.31 (1H, d, $J = 8.0$ Hz), 7.45 (1H, d, $J = 7.7$ Hz), 10.06 (1H, bs). ^{13}C NMR (100 MHz, DMSO-d_6, 116 °C): δ 12.9, 26.8, 27.3, 27.5, 30.7, 38.3, 54.9, 59.4, 77.7, 105.0, 108.7, 110.0, 110.5, 117.4, 117.5, 119.7, 128.9, 134.4, 140.3, 145.9, 154.2, 171.5. IR (NaCl neat) 3376, 3083, 3057, 2976, 2933, 1697, 1503, 1462, 1376, 1167, 1021, 917, 861, 742 cm^{-1}. HRMS (FAB+): Calcd for $C_{23}H_{32}N_2O_4$: 400.236208. Found 400.236330 (M^+).

***N*-[(1,1-Dimethylethoxy)carbonyl]-2-(1,1-dimethyl-2-propenyl)-D,L-tryptophan (5).** *N*-[(1,1-Dimethylethoxy)carbonyl]-2-(1,1-dimethyl-2-propenyl)-D,L-tryptophan ethyl ester (1.97 g, 4.60 mmol) was stirred with LiOH (589 mg, 25 mmol) in a THF:H_2O solution (2:1) (16 mL) overnight. The solution was acidified with 10% $KHSO_4$(aq) and extracted three times with EtOAc. The organic layer was dried over anhydrous Na_2SO_4, and the solvent was concentrated under reduced pressure to afford 1.63 g (89%) of **5** as an amorphous solid. The product was deemed sufficiently pure to use directly for the next step without further purification. ^{1}H NMR (300 MHz, DMSO-d_6, 120 °C): δ 1.28 (9H, s), 1.55 (3H, s), 1.56 (3H, s), 3.09 (1H, dd, $J = 8.1, 14.7$ Hz), 3.46 (1H, dd, $J = 6.2, 14.7$), 4.30 (1H, dd, $J = 8.1, 14.7$ Hz), 5.06 (1H, dd, $J = 1.1, 10.6$ Hz), 5.10 (1H, dd, $J = 0, 17.2$ Hz), 5.98 (1H, d, $J = 5.9$ Hz), 6.22 (1H, dd, $J = 10.6, 17.2$ Hz), 6.92 (1H, ddd, $J = 1.1, 7.7, 7.7$ Hz), 7.00 (1H, ddd, $J = 0.8, 7.7, 7.7$ Hz), 7.31 (1H, d, $J = 7.7$ Hz), 7.53 (1H, d, $J = 7.7$ Hz), 10.02 (1H, bs), 11.51 (1H, very bs). ^{13}C NMR (100 MHz, DMSO-d_6, 120 °C): δ 27.1, 27.3, 27.4, 38.3, 54.6, 77.6, 105.4, 110.1, 110.4, 117.5, 119.6, 129.0, 134.4, 140.3, 146.0, 154.2, 172.7. IR (NaCl neat) 3368−2563, 3368, 3087, 3053, 2974, 2926, 1712, 1502, 1460, 1394, 1367, 1245, 1164, 1054, 1010, 919, 742 cm $^{-1}$. HRMS (FAB+): Calcd for $C_{21}H_{28}N_2O_4$: 372.2049. Found 372.2052 (M^+).

Synthesis of 6a. *N*-[(1,1-Dimethyethoxy)carbonyl]-*N*-(3-oxobutyl)-glycine Ethyl Ester. The hydrochloride salt of glycine ethyl ester was neutralized by the addition of 1 equiv of aqueous 10% Na_2CO_3 and extracting five times with CH_2Cl_2. After drying the organic layer over anhydrous Na_2SO_4, the solvent was removed under reduced pressure, and the crude free amine was obtained. Glycine ethyl ester (9.06 g, 87.8 mmol) was stirred with methyl vinyl ketone (7.28 mL, 1.0 equiv) in acetonitrile (88 mL) at room temperature under argon in the absence of light. After 3 h, the solvent was removed under reduced pressure, and the flask was placed under vacuum for 1 h. The free amine decomposes fairly rapidly upon standing, and it was found best to proceed directly to the next step without further purification. To a solution of the adduct obtained above (13.76 g, 79.4 mmol) at 0 °C in dioxane (160 mL) were added di-*tert*-butyl pyrocarbonate (17.3 g, 1.0 equiv, 79.4 mmol), 1 M NaOH solution (79.4 mL), and deionized water (79.4 mL). The reaction was allowed to stir under argon, in the absence of light, at room-temperature overnight. The reaction was worked up by adding saturated NaCl solution (100 mL), extracting three times with EtOAc, drying over anhydrous Na_2SO_4, and evaporating of the solvent under reduced pressure. The crude product was purified by Kugelrohr distillation; the product distilled at 102 °C at 1 mmHg affording 15.64 g of the product as an oil (65% for the two steps). ^{1}H NMR (300 MHz, DMSO-d_6, 120 °C): δ 1.23 (3H, ddd, $J = 1.6, 7.3, 7.3$ Hz), 1.41 (9H, s), 2.10 (3H, s), 2.70 (2H, t, $J = 6.6$ Hz), 3.44 (2H, ddd, $J = 1.5, 6.6, 6.6$ Hz), 3.92 (2H, s), 4.14 (2H, dddd, $J = 1.5, 7.0, 7.0, 7.0$ Hz). ^{13}C NMR (100 MHz, DMSO-d_6, 120 °C): 13.3, 27.4, 27.6, 28.9, 41.6, 42.8, 49.0, 59.6, 78.8, 154.0, 169.1, 205.9. IR (NaCl, neat): 3611, 3398, 2978, 2936, 1748, 1698, 1462, 1397, 1367, 1250, 1162, 1129, 1029, 894, 866, 778 cm^{-1}. HRMS (FAB+) Calcd. for $C_{13}H_{24}NO_5$: 274.165448. Found 274.165921 (M + H).

3-Hydroxy-3-methyl-1,2-pyrrolidinedicarboxylic Acid 1-(1,1-Dimethylethyl) 2-Ethyl Ester (6a). A solution of the N-Boc-protected compound obtained above (5 g, 18.29 mmol) in toluene (100 mL) was cooled to 0 °C. Solid potassium *tert*-butoxide (2.05 g, 1.0 equiv) was added portionwise, and the solution was stirred under argon for 45 min at 0 °C. The reaction was quenched by the addition of ice cold 10% aqueous $KHSO_4$ (pH = 2−3). The organic layer was separated from the aqueous layer, and the aqueous layer was extracted three times with CH_2Cl_2. The combined organic fractions were washed with pH = 7 phosphate buffer and brine successively. The organic layer was dried over anhydrous Na_2SO_4 and evaporated to give a yellowish oil. The oil was resuspended in CH_2Cl_2 and extracted three times with pH = 10 Na_2CO_3 buffer. The combined aqueous extracts were extracted two more times with CH_2Cl_2. The organic layers were washed with brine, dried over anhydrous Na_2SO_4, and evaporated to give 1.89 g (38%) of the product as an off-white amorphous solid. ^{1}H NMR (300 MHz, DMSO-d_6, 120 °C): δ 1.23 (3H, ddd, $J = 1.0, 7.0, 7.0$ Hz), 1.40 (12H, s), 1.77 (1H, m), 1.98 (1H, m), 3.36 (1H, m), 3.48 (1H, m), 3.89 (1H, d, $J = 2.2$ Hz), 4.12 (2H, dddd, $J = 2.2, 7.0, 7.0, 7.0$ Hz), 4.62 (1H, bs). ^{13}C NMR (100 MHz, DMSO-d_6, 116 °C): δ 13.5, 26.6, 27.4, 37.6, 43.8, 59.1, 68.4, 78.2, 152.8, 168.9. IR (NaCl, neat•): 3446, 3093, 2977, 2935,m 2900, 1743, 1681, 1456, 1403, 1367, 1161, 1097, 1033, 926, 860, 774, 739 cm[1]. HRMS (FAB+): Calcd for $C_{13}H_{24}N_1O_5$: 274.165448. Found 274.166420 (M + H).

1-[*N*-[(1,1-Dimethylethoxy)carbonyl]-2-(1,1-dimethyl-2-propenyl)-D,L-tryptophyl]-3-hydroxy-3-methyl-D,L-proline Ethyl Ester (7). Compound **6a** (1.21 g, 4.43 mmol) was stirred with TFA (6.8 mL) in CH_2Cl_2 (7 mL) at 0 °C. The reaction was allowed to come to room temperature and stir for an additional 3 h. A saturated solution of $NaHCO_3$ was added until the solution became basic, and the organic layer was separated from the aqueous phase. The aqueous layer was extracted three more times with EtOAc, the combined organic layers were dried over anhydrous Na_2SO_4, and the solvent was removed under reduced pressure. Compound **6b** (*β*-hydroxy-*β*-methylproline ethyl ester) was mixed with compound **5** (1.65 g, 4.43 mmol), BOP reagent (1.96 g, 4.43 mmol), and Et_3N (1.24 mL) in acetonitrile (67 mL) at room temperature for 4 h. A saturated aqueous solution of NaCl was added, and the reaction was extracted four times with EtOAc. The combined organic layers were washed with 2 M HCl, water, 10% $NaHCO_3$ (aq), water, and brine successively. The organic phase was dried over anhydrous Na_2SO_4 and evaporated to dryness under reduced pressure. The product was partially purified by flash silica gel column chromatography using 4% MeOH/CH_2Cl_2; the slightly impure mixture of diastereomers **7** (1.88 g, 80%) were directly carried on to the next step without further purification.

3-[[2-(1,1-Dimethyl-2-propenyl)-1*H*-indol-3-yl]methyl]hexahydro-8-hydroxy-8-methylpyrrolo[1,2-*a*]pyrazine-1,4-dione (8). To a solution of **7** (527 mg, 1.00 mmol) in CH_2Cl_2 (2 mL) at 0 °C was added TFA (1.6 mL). The ice bath was removed, and the mixture was allowed to come to room temperature and stir for an additional 3 h. A saturated solution of $NaHCO_3$ was added until the solution became basic, and the organic layer was separated from the aqueous phase. The aqueous layer was extracted three more times with EtOAc, the combined organic layers were dried over anhydrous Na_2SO_4, and the solvent was removed under reduced pressure. The crude free amine was then dissolved in toluene (5 mL) with 2-hydroxypyridine (19 mg), the solution was refluxed overnight under argon, and the solvent was removed under reduced pressure. The four diastereomers could be partially separated

using PTLC, but in practice the mixture of products was purified as a mixture of diasteromers via radial silica gel chromatography using an elutant of 2% MeOH/CH_2Cl_2 to afford 361 mg (95%) of **8** as a mixture of diastereomers (solid). Data for two of the diastereomers, **8a** and **8d**, are descibed below (relative stereochemistry not assigned); diastereomers **8b** and **8c** could not be separated.

8a: ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.57 (6H, s), 1.62 (3 H, s), 1.89 (1H, m), 2.18 (1H, ddd, J = 7.3, 7.3, 13.5 Hz), 2.95 (1H, bs), 3.21, (1H, dd J = 9.7, 15.4 Hz), 3.74 (3H, m), 3.91 (1H, d, J = 1.5 Hz), 4.40 (1H, dd, J = 2.2, 11.3 Hz), 5.19 (1H, dd, J = 0, 17.2 Hz), 5.20 (1H, dd, J = 0, 11.0 Hz), 5.81 (1H, bs), 6.15 (1H, dd, J = 10.9, 16.8 Hz), 7.12 (1H, ddd, J = 1.1, 7.3, 7.3 Hz), 7.19 (1H, ddd, J = 1.5, 7.3, 7.3 Hz), 7.34 (1H, d, J = 8.0 Hz), 7.50 (1H, d, J = 7.7 Hz), 8.10 (1H, bs). ^{13}C NMR (75 MHz, $CDCl_3$): δ 25.9, 26.1, 27.8, 27.9, 36.9, 39.0, 43.3, 54.5, 65.8, 77.8, 104.5, 110.9, 112.9, 117.8, 120.1, 122.2, 128.0, 132.2, 141.5, 145.6, 166.0, 168.0. IR (NaCl neat) 3360, 3054, 2968, 2924, 1666, 1651, 1462, 1434, 1302, 1262, 1138, 1105, 1010, 919, 734 cm $^{-1}$. HRMS (FAB+): Calcd. for $C_{22}H_{28}N_3O_3$: 382.213067. Found 382.212574 (M + H). R_f 0.75 (eluted twice with 2% MeOH/CH_2Cl_2).

8d: ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.54 (3 H, s), 1.55 (3H, s), 2.04 (2H, m), 2.32 (1H, bs), 2.66 (3H, d, J = 9.2 Hz), 3.25 (1H, dd, J = 9.5, 14.3), 3.46 (1H, dd, J = 14.6, 3.6), 3.53 (2H, m), 3.74 (1H, m), 4.25 (1H, m), 5.16 (1H, dd, J = 1.1, 10.6 Hz), 5.19 (1H, dd, J = 0.8, 17.2 Hz), 6.12 (1H, dd, J = 10.6, 17.6 Hz), 6.18 (1H, bs), 7.12 (2H, m), 7.28 (1H, dd, J = 1.5, 6.6 Hz), 7.53 (1H, dd, J = 1.5, 7.0 Hz), 8.09 (1H, bs). ^{13}C NMR (300 MHz, $CDCl_3$): δ 25.7, 27.7, 28.0, 36.6, 36.9, 39.1, 43.3, 58.2, 65.0, 77.9, 105.1, 110.5, 111.9, 118.4, 119.8, 121.9, 128.9, 134.2, 141.4, 146.0, 166.2, 167.6. IR (NaCl neat) 3340, 3084, 3044, 2970, 2924, 1671, 1658, 1461, 1447, 1372, 1327, 1198, 1138, 1009, 987, 732 cm $^{-1}$. HRMS (FAB+): Calcd for $C_{22}H_{28}N_3O_3$: 382.213067. Found 382.211498 (M + H). R_f 0.43 (eluted twice with 2% MeOH/CH_2Cl_2).

(3*R*,*S*)-3-[[2-(1,1-Dimethyl-2-propenyl)-1*H*-indol-3-yl]methyl]-2,3,6,7-tetrahydro-8-methylpyrrolo[1,2-*a*]pyrazine-1,4-dione (9). Compound **8** (535 mg, 1.40 mmol) was cooled to 0 °C in THF (5.6 mL) under an argon atmosphere. Pyridine (226 μL, 2.0 equiv) was added, and the solution was stirred for ~15 min. $SOCl_2$ (112 μL, 1.1 equiv) was added, and the mixture was allowed to come to room temperature over 3 h. Water was added to the reaction mixture which was then extracted three times with EtOAc. The organic layer was dried over anhydrous Na_2SO_4 and evaporated to dryness under reduced pressure. The product was purified by flash silica gel column chromatography using 2% MeOH/CH_2Cl_2 to afford 381 mg (75%) of compound **9** as a glass. ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.56 (6H, s), 2.02 (3H, s), 2.69 (2H, dd, J = 9.2, 9.2 Hz), 3.21 (1H, dd, J = 11.3, 14.6 Hz), 3.72 (1 H, dd J = 3.3, 14.7 Hz), 3.93 (2H, ddd, J = 3.0, 11.7, 11.7 Hz), 4.47 (1H, d, J = 10.6 Hz), 5.17 (1H, dd, J = 0, 10.26 Hz), 5.18 (1H, dd, J = 0, 17.2 Hz), 5.55 (1H, bs), 6.13 (1H, dd, J = 10.6, 17.6 Hz), 7.11 (1H, ddd, J = 1.1, 7.3, 7.3 Hz), 7.18 (1H, ddd, J = 0.7, 7.3, 7.3 Hz), 7.32 (1H, d, J = 7.7 Hz), 7.55 (1H, d, J = 7.7 Hz), 8.07 (1H, bs). ^{13}C NMR (75 MHz, $CDCl_3$): δ 13.7, 26.8, 28.0, 30.8, 33.9, 39.1, 43.1, 57.0, 104.8, 110.7, 112.4, 118.3, 120.0, 122.0, 128.9, 134.30, 134.33, 141.5, 145.7, 158.1, 162.2. IR (NaCl neat) 3344, 3047, 2968, 1682, 1645, 1440, 1324, 1251, 1114, 1007, 917, 744 cm^{-1}. HRMS (FAB+): Calcd for $C_{22}H_{26}N_3O_2$: 364.2025. Found 364.2032 (M + H). R_f 0.2 (eluted with 2% MeOH/CH_2Cl_2).

(3*R*,*S*)-3-[[2-(1,1-Dimethyl-2-propenyl)-1*H*-indol-3-yl]methyl]-6,7-dihydro-1-methoxy-8-methylpyrrolo[1,2-*a*]pyrazin-4(3*H*)-one (10). A solution of **9** (257 mg, 0.7 mmol) was stirred with $(CH_3)_3OBF_4$ (314 mg, 2.12 mmol, 3.0 equiv) and anhydrous K_2CO_3 (5 equiv, 489 mg, 3.54 mmol, 5.0 equiv) in CH_2Cl_2 (7 mL) for 7 h at ambient temperature under an argon atmosphere. The reaction was poured into ice water and extracted with CH_2Cl_2 three times. The combined organic layers were washed with brine, dried over anhydrous Na_2SO_4, concentrated under reduced pressure, and purified by silica gel column chromatography (eluted with 50% EtOAc/hexanes) to yield 192 mg (72%) of the azadiene **10** as a brittle foam. R_f 0.4 (eluted with 50% EtOAc/hexanes). ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.61 (3H, s), 1.62 (3H, s), 1.99 (3H, s), 2.55 (2H, m), 3.09 (1H, dd, J = 9.2, 14.3 Hz), 3.66 (3H, s), 3.79 (3H, m), 4.59 (1H, d, J = 7.0 Hz), 5.15 (1H, dd J = 1.1, 10.3 Hz), 5.18 (1H, dd J = 1.1, 17.2 Hz), 6.15 (1H, dd, J = 10.3, 17.2 Hz), 7.04 (1H, ddd, J = 1.1, 8.1, 8.1 Hz), 7.15 (1H, ddd, J = 1.1, 7.0, 7.0 Hz), 7.26 (1H, d, J = 8.1 Hz), 7.65 (1H, d, J = 7.7 Hz), 7.86 (1H, bs). ^{13}C NMR (75 MHz, $CDCl_3$): δ 13.7, 27.7, 27.8, 31.4, 34.0, 39.3, 42.8, 52.7, 64.0, 108.1, 109.9, 111.8, 118.6, 119.7, 121.1, 122.8, 124.5, 130.3, 134.1, 140.0, 146.2, 152.6, 166.3. IR (NaCl neat) 3345, 2962,2924, 1676, 1634, 1456, 1335, 1304, 1242, 1051, 917, 741 cm^{-1}. HRMS (FAB+): Calcd. for $C_{23}H_{28}N_3O_2$: 378.217697. Found 378.218152 (M + H).

2,3,11,12,12a,13-Hexahydro-14-methoxy-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(nitrilometheno)-1*H*-indolizino[7,6-*b*]carbazol-5-one (12−15). Azadiene **10** (264 mg, 0.70 mmol) was stirred in MeOH (47 mL) and 20% KOH (aq) (12.6 mL) under an argon atmosphere at 0 °C. The reaction mixture was allowed to come to room temperature and continued to stir for 10 h. When the reaction was complete as indicated by TLC analysis, phosphate buffer (pH = 7) was added until the solution was neutral. The aqueous phase was extracted three times with EtOAc. The combined organic extracts were washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. The mixture of diastereomers could be partially separated by flash silica gel column chromatography (eluted with 2−4% MeOH/CH_2Cl_2); however, cycloadducts **13** and **14** had to be separated by successive PTLC (eluted with 2% MeOH/CH_2Cl_2). The order of elution was (from fastest mobility to slowest mobility): **15**, **14**, **13**, and finally **12**. Yield: **15**, 23 mg; **14**, 37 mg; **13**, 62 mg; **12**, 85 mg (78% combined yield). Data for each is as follows:

(1S,5a*R*,12a*R*,13a*S*)-*rel*-2,3,11,12,12a,13-Hexahydro-14-methoxy-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(nitrilometheno)-1*H*-indolizino[7,6-*b*]carbazol-5-one (12). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.08 (3H, s), 1.22 (3H, d, J = 7.0 Hz), 1.31 (3H, s), 1.64−1.78 (3H, m), 2.16 (1H, m), 2.28 (1H, dd, J = 9.2, 5.5 Hz), 2.90 (1H, m), 3.12 (1H, d, J = 15.7 Hz); 3.26 (1H, m), 3.56 (1H, m), 3.81 (3H, s), 4.03 (1H, d, J = 15.7 Hz), 7.12 (2H, m), 7.27 (1H, d, J = 7 Hz), 7.57 (1H, d, J = 7 Hz), 7.74 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.02, 22.1, 26.6, 27.7, 28.6, 32.4, 34.3, 35.2, 42.4, 46.4, 54.4, 65.7, 66.0, 106.8, 110.2, 118.8, 118.9, 121.2, 127.6, 136.5, 139.5, 171.1, 172.8. IR (NaCl, neat): 3306, 2969, 1668, 1651, 1455, 1428, 1345, 1310, 1194, 995, 740, 714 cm^{-1}. HRMS (FAB+) Calcd for $C_{23}H_{28}N_3O_2$: 378.2181. Found 378.2176 (M + H). R_f 0.40 (eluted twice with 2% MeOH/CH_2Cl_2).

(1*R*,5a*R*,12a*R*,13a*S*)-*rel*-2,3,11,12,12a,13-Hexahydro-14-methoxy-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(nitrilometheno)-1*H*-indolizino[7,6-*b*]carbazol-5-one (13). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.02 (3H, s), 1.26 (3H, s), 1.43 (3H, d, J = 7.0 Hz), 1.73 (1H, m), 1.86 (1H, dd, J = 10.6, 12.5 Hz), 2.11(1H, dd, J = 4.8, 12.5 Hz), 2.11 (1H, m), 2.29 (1H, dd, J = 10.6, 4.8 Hz), 2.38 (1H, m), 3.12 (1H, d, J = 16.1 Hz); 3.25 (1H, m), 3.62 (1H, m), 3.65 (3H, s), 4.06 (1H, d, J = 16.1 Hz), 7.15 (2H, m), 7.31 (1H, d, J = 7 Hz), 7.61 (1H, d, J = 7 Hz), 7.73 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.9, 22.9, 27.8, 28.4, 32.3, 32.9, 35.0, 41.1, 42.8, 47.5, 54.0, 65.8, 66.2, 106.9, 110.3, 118.8, 119.0, 121.9, 127.6, 136.5, 139.6, 172.9, 173.1. IR (NaCl, neat): 3306, 3047, 2951, 1167, 1633, 1462, 1435, 1372, 1311, 1185, 980, 743 cm^{-1}. HRMS (FAB+) Calcd for $C_{23}H_{28}N_3O_2$: 378.2181. Found 378.2163 (M + H). R_f 0.49 (eluted twice with 2% MeOH/CH_2Cl_2).

(1*S*,5a*R*,12a*S*,13a*S*)-*rel*-2,3,11,12,12a,13-Hexahydro-14-methoxy-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(nitrilometheno)-1*H*-indolizino[7,6-*b*]carbazol-5-one (14). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.13 (3H, s), 1.19 (3H, d, J = 7.0 Hz), 1.25 (3H, s), 1.68 (1H, m), 1.88 (1H, dd, J = 9.8, 12.9 Hz), 1.97 (1H, dd, J = 3.9, 12.9 Hz), 2.12 (1H, m), 2.23 (1H, dd, J = 9.8, 3.9 Hz), 2.88 (1H, m), 3.22 (1H, m), 3.29 (1H, d, J = 17.2 Hz), 3.65 (3H, s), 3.68 (1H, m), 3.92 (1H, d, J = 17.2 Hz), 7.09 (2H, m), 7.27 (1H, d, J = 7 Hz), 7.57 (1H, d, J = 7 Hz), 7.94 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.1, 25.2, 25.9, 27.9, 28.4, 32.0, 34.2, 35.0, 42.5, 45.0, 54.2, 65.8, 66.9, 106.1, 110.3, 118.8, 119.4, 121.3, 128.0, 136.4, 139.8, 170.8, 172.1. IR (NaCl, neat, cm^{-1}): 3407, 3312, 3053, 2964, 1667, 1455, 1427, 1345, 1306, 1230, 1180, 1042, 995, 815, 738. HRMS (FAB+) Calcd for $C_{23}H_{28}N_3O_2$: 378.2181. Found 377.2109 (M − H). R_f 0.55 (eluted twice with 2% MeOH/CH_2Cl_2).

(1*R*,5a*R*,12a*S*,13a*S*)-*rel*-2,3,11,12,12a,13-hexahydro-14-methoxy-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(nitrilometheno)-1*H*-indolizino[7,6-*b*]carbazol-5-one (15). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.15 (3H, s), 1.25 (3H, s), 1.41 (3H, d, J = 7.0 Hz), 1.71 (1H, m), 1.86 (1H, dd, J = 9.9, 13.2 Hz), 1.97 (1H, dd, J = 4.4, 13.2 Hz), 2.10 (1H, m), 2.33 (1H, m), 2.38 (1H, dd, J = 9.9, 4.4 Hz), 3.27 (1H, d, J = 17.2 Hz), 3.35 (1H, m), 3.57 (1H, m), 3.65 (3H, s), 3.88 (1H, d, J = 17.2 Hz), 7.10 (2H, m), 7.28 (1H, d, J = 7 Hz), 7.59 (1H, d, J = 7 Hz), 7.70 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.2, 25.3, 26.1, 28.4, 32.9, 33.6, 34.9, 40.7, 43.0, 45.8, 53.8, 66.2, 66.7, 106.4, 110.3, 118.9, 119.2, 121.5, 128.0, 136.4, 139.7, 171.1, 173.4. IR (NaCl, neat): 3407, 3310, 2958, 2918, 1660, 1626, 1461, 1423, 1307, 1283, 1008, 741 cm^{-1}. HRMS (FAB+) Calcd for $C_{23}H_{28}N_3O_2$: 378.2181. Found 378.2173, (M + H). R_f 0.57 (eluted twice with 2% MeOH/CH_2Cl_2).

General Procedure for Lactim Ether Deprotection of Cyclo Adducts (12−15). One equivalent of the lactim ether cycloadduct was stirred in THF (0.025 M) at 0 °C. To this solution was added 0.1 M HCl (3.0 equiv), and the reaction was stirred 5−15 min until starting material was no longer detected by TLC analysis. The reaction was netralized with pH = 7 phosphate buffer and extracted three times with EtOAc. The combined organic layers were dried over anhydrous Na_2SO_4, and the solvent was removed under reduced pressure. In the case of cycloadducts **13** and **15**, the ring opened products **17** and **22** were obtained. In the case of cycloadduct **12**, a small percentage of conversion to the corresponding ring-opened amine methyl ester was sometimes observed. These ring-opened amine methyl esters were recyclized by refluxing in toluene (0.025 M) overnight. The corresponding piperazinedione products (**16**, **18**, **20**, and **23**) were purified using flash silica gel column chromatography (eluted with 2−4% MeOH/CH_2Cl_2). Data for each is as follows:

(1*S*,5a*R*,12a*R*,13a*S*)-*rel*-2,3,11,12,12a,13-hexahydro-1,12,12-trimethyl-5*H*,6H-5a,13a-(iminomethano)-1*H*-indolizino[7,6-*b*]carbazole-5,14-dione (16) from 12. Yield: 16.5 mg (85%). In this instance, a small percentage of the ring-opened amine methyl ester was observed by ^{1}H NMR analysis and TLC. The mixture of ring-opened product and the desired piperazinedione was refluxed overnight in toluene and purifed by PTLC. ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.08 (3H, s), 1.20 (3H, d, J = 6.6 Hz), 1.33 (3H, s), 1.67 (1H, m), 1.91 (1H, dd, J = 5.0, 13.4 Hz), 2.03 (1H, dd, J = 10.1, 13.4 Hz), 2.16 (1H, m), 2.25 (1H, dd, J = 3.9, 10.1 Hz), 2.60 (1H, d, J = 15.4 Hz), 3.01 (1H, m), 3.28 (1H, m), 3.58 (1H, m), 3.87 (1H, d, J = 15.4 Hz) 5.84 (1H, bs), 7.11 (2H, m), 7.27 (1H, d, J = 7.3 Hz), 7.50 (1H, d, J = 7.7 Hz), 7.72 (1H, bs). ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.8, 21.3, 23.8, 24.8, 28.1, 31.3, 34.2, 34.7, 42.5, 48.2, 59.2, 66.9, 103.4, 110.7, 117.5, 118.2, 120.6, 126.5, 136.5, 140.7, 168.3, 173.4. IR (NaCl, neat): 3313, 3066, 2960, 2913, 1681, 1455, 1404, 1257, 1187, 1088, 1022, 795, 734, 693 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{25}N_3O_2$: 363.1946. Found 363.1943 (M^+). R_f 0.40 (eluted twice with 4% MeOH/CH_2Cl_2).

(1*R*,5a*R*,12a*R*,13a*S*)-*rel*-2,3,11,12,12a,13-Hexahydro-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(iminomethano)-1*H*-indolizino[7,6-*b*]carbazole-5,14-dione (18) from 13 via 17. Yield: 26 mg (63%). Data for **17**: ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.95 (3H, d, J = 6.6 Hz), 1.28 (1H, d, J = 13.7 Hz), 1.31 (1H, d, J = 13.3 Hz), 1.39 (3H, s), 1.53 (1H, m), 1.56 (3H, s), 1.76 (1H, d, J = 13.3 Hz), 2.00 (3H, m), 2.82 (1H, d, J = 16.4 Hz), 2.88 (1H, d, J = 13.7 Hz), 2.93 (1H, d, J = 16.4 Hz), 3.68 (2H, m), 3.74 (3H, s), 7.02 (1H, m), 7.08 (1H, m), 7.25 (1H, d, J = 7.8 Hz), 7.35 (1H, d, J = 7.8 Hz), 8.11 (1H, s). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.2, 28.4, 29.4, 31.5, 31.9, 33.2, 33.9, 45.2, 45.4, 47.5, 52.3, 58.2, 71.3, 103.5, 110.6, 118.1, 119.3, 121.5, 127.6, 136.3, 138.9, 172.2, 174.4. IR (NaCl, neat): 3352, 2964, 1725, 1681, 1455, 1392, 1262, 1115, 1020, 808, 761, 732 cm^{-1}. HRMS (FAB+) Calcd for $C_{23}H_{30}N_3O_3$: 396.2287. Found 396.2281 (M + H). R_f 0.30 (eluted twice with 4% MeOH/CH_2Cl_2). The ring-opened amine methyl ester **17** was cyclized to the piperazinedione **18** as described above. Data for **18**: ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.07 (3H, s), 1.29 (3H, s). 1.52 (3H, d, J = 7 Hz), 1.80 (1H,dd, J = 5.0, 13.5 Hz), 1.85 (1H, m), 2.09 (1H, m), 2.30 (1H, m), 2.41 (1H, dd, J = 10.5, 13.5 Hz), 2.58 (1H, dd, J = 5.0, 10.5 Hz), 2.59 (1H, d, J = 15.2 Hz), 3.25 (1H, m), 3.65 (1H, m), 3.83 (1H, d, J = 15.2 Hz), 5.95 (1H, bs), 7.08 (1H, ddd, J = 1.2, 7.8, 7.8 Hz), 7.13 (1H, ddd, J = 0.8, 7.8, 7.8 Hz), 7.26 (1H, d, J = 7.8 Hz), 7.48 (1H, d, J = 7.8 Hz), 7.78 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$ + 1 drop DMSO-d_6): δ 12.9, 22.2, 24.5, 28.0, 29.4, 31.0, 34.5, 41.5, 43.0, 49.3, 59.8, 66.6, 103.9, 110.5, 118.6, 119.7, 121.0, 126.7, 136.4, 139.9, 169.9, 173.5. IR (NaCl, neat): 3664, 3326, 2960, 1677, 1453, 1258, 1092, 799, 703 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{26}N_3O_2$: 364.2025. Found 364.2023 (M + H). R_f 0.30 (eluted twice with 4% MeOH/CH_2Cl_2).

(1*S*,5a*R*,12a*S*,13a*S*)-*rel*-2,3,11,12,12a,13-Hexahydro-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(iminomethano)-1*H*-indolizino[7,6-*b*]carbazole-5,14-dione (20) from cycloadduct 14. Yield: 14.5 mg, (100%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.17 (3H, d, J = 7.0 Hz), 1.26 (3H, s), 1.33 (3H, s), 1.66 (1H, m), 1.85 (1H, dd, J = 3.9, 13.7 Hz), 2.02 (1H, dd, J = 10.1, 13.3 Hz), 2.14 (1H, m), 2.25 (1H, dd, J = 3.9, 10.1), 2.89 (1H, d, J = 17.9 Hz), 2.98 (1H, m), 3.26 (1H, m) 3.73 (1H, m), 3.90 (1H, d, J = 17.6 Hz), 5.90 (1H, bs), 7.09 (1H, m), 7.15 (1H, m), 7.30 (1H, d, J = 8 Hz), 7.50 (1H, d, J = 7.8 Hz), 7.89 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.3, 23.8, 25.2, 27.3, 29.0, 31.9, 34.5, 34.6, 42.9, 44.9, 61.2, 68.2, 103.7, 110.7, 118.4, 119.7, 122.1, 127.2, 136.4, 139.6, 169.2, 173.1. IR (NaCl, neat): 3298, 2962, 1682, 1455, 1399, 1302, 1231, 742 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{25}N_3O_2$: 363.1946. Found 363.1949 (M^+). R_f 0.40 (eluted twice with 4% MeOH/CH_2Cl_2).

(1*R*,5a*R*,12a*S*,13a*S*)-*rel*-2,3,11,12,12a,13-Hexahydro-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(iminomethano)-1*H*-indolizino[7,6-*b*]carbazole-5,14-dione (23) from Cycloadduct 15 via 22. Yield: 4.7 mg, (49%). Data for **22**: ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.04 (3H, d, J = 7.0 Hz), 1.34 (3H, s), 1.46 (3H, s), 1.58 (2H, bs), 1.66 (1H, m), 1.90 (1H, dd, J = 9.8, 14.1 Hz), 2.02 (1H, m), 2.05 (1H, dd, J = 9.4, 9.4), 2.18 (1H, m), 3.03 (1H, dd, J = 9.0, 14.1 Hz), 3.13 (1H, d, J = 16.4 Hz), 3.15 (1H, d, J = 16.4 Hz), 3.64 (3H, s), 3.65 (1H, m), 3.73 (1H, m), 7.08 (1H, ddd, J = 7.8, 7.8, 1.2), 7.14 (1H, ddd, J = 7.8, 7.8, 1.2), 7.30 (1H, d, J = 7.8 Hz), 7.52 (1H, d, J = 7.8 Hz), 7.84 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.3, 25.3, 28.2, 29.2, 30.7, 34.1, 35.0, 45.1, 45.2, 47.6, 51.9, 57.1, 68.5, 105.4, 110.5, 118.5, 119.4, 121.8, 128.1, 136.5, 139.8, 172.4, 173.3. IR (NaCl, neat): 3301, 2961, 1732, 1637, 1451, 1220, 735 cm^{-1}. HRMS (FAB+) Calcd for $C_{23}H_{30}N_3O_3$: 396.2287. Found 396.2282 (M + H). R_f 0.6 (eluted with 4% MeOH/CH_2Cl_2). The ring-opened amine methyl ester **22** was cyclized to the piperazinedione **23** as described above**.** Data for **23**: ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.29 (3H, s), 1.32 (3H, s), 1.50 (3H, d, J = 7.0 Hz), 1.82 (1H, m), 1.99 (1H, dd, J = 10.5, 13.6 Hz), 2.10 (1H, m), 2.22 (1H, dd, J = 3.5, 13.6 Hz), 2.31 (2H, m), 2.85 (1H, d, J = 17.9 Hz), 3.44 (1H, m), 3.61 (1H, m), 3.87 (1H, d J = 17.9 Hz), 5.73 (1H, bs), 7.10 (1H, t, J = 7.4 Hz), 7.16 (1H, t, J = 7.8 Hz), 7.30 (1H, d, J = 7.8 Hz), 7.50 (1H, d, J = 7.4 Hz), 7.85 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.6, 23.9, 25.4, 29.0, 32.5, 32.8, 34.5, 41.1, 43.3, 45.7, 61.2, 67.7, 103.7, 110.7, 118.4, 119.7, 122.2, 127.1, 136.4, 139.6, 170.2, 172.8. IR (NaCl, neat): 3228, 2925, 1684, 1670, 1570, 1453, 1406, 1291, 871, 737 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{25}N_3O_2$: 363.1946. Found 363.1953 (M^+). R_f 0.5 (eluted with 4% MeOH/CH_2Cl_2).

Racemic VM55599 and Diastereoisomers (18, 19, 21). General Procedure for DIBAH Reduction of 16, 18, 20, and 23. The cycloadduct (**16**, **18**, **20**, or **23**) (0.005 M in toluene) was stirred at 0 °C under an atmosphere of argon, and DIBAH (20 equiv as a 1.0 M solution in toluene) was added. The reaction was allowed to come to room temperature and stirred for 24 h. The reaction was again cooled to 0 °C and $Na_2SO_4 \cdot 10H_2O$ was added slowly until bubbling subsided. The mixture was stirred an additional 30 min and then filtered through a fritted funnel. The solid residue was rinsed with ethyl acetate, and the combined filtrates were evaporated under reduced pressure. The product was isolated via flash silica gel column chromatography or PTLC using 2% MeOH/CH_2Cl_2.

(±)-VM55599. Yield: 13.5 mg, (86%); obtained as an amorphous powder. ^{1}H NMR (400 MHz, $CDCl_3$ + 1 drop DMSO-d_6): δ 1.00 (3H, d, J = 7.0 Hz), 1.31 (3H, s), 1.37 (1H, m), 1.39 (3H, s), 1.73 (1H, dd, J = 11.7, 13.2 Hz), 1.96 (1H, dd, J = 4.3, 13.2 Hz), 2.13 (3H, m), 2.24 (1H, dd, J = 1.6, 10.1 Hz), 2.76 (1H, d, J = 15.2 Hz), 2.90 (1H, d, J = 15.2 Hz), 2.96 (2H, m), 3.45 (1H, d, J = 10.1 Hz), 6.28 (1H, bs), 7.04 (1H, ddd, J = 0.8, 7.8, 7.8 Hz), 7.11 (1H, ddd, J = 1.2, 7.0, 7.0 Hz), 7.29 (1H, d, J = 7.3 Hz), 7.39 (1H, d, J = 7.8 Hz), 8.40 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$ + 1 drops DMSO-d_6): δ 17.5, 24.0, 26.8, 30.1, 30.2, 30.5, 33.0, 34.2, 46.6, 53.6, 55.7,

58.9, 66.4, 104.1, 110.6, 117.7, 119.2, 121.5, 126.8, 136.4, 141.1, 174.8. I. R. (NaCl, neat): 3303, 3048, 2920, 1650, 1454, 1296, 779, 734, 695 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{28}N_3O$: 350.2232. Found 350.2235 (M + H). R_f 0.38 (eluted with 4% MeOH/CH_2Cl_2). This synthetic compound was identical to natural VM55599 (obtained from *Penicillium* sp. IMI332995) by TLC (silica gel, eluted with 4% MeOH/CH_2Cl_2), ^{1}H NMR, and ^{13}C NMR.

(1*R*,5a*R*,12a*R*,13a*S*)-*rel*-2,3,11,12,12a,13-Hexahydro-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(iminomethano)-1*H*-indolizino[7,6-*b*]carbazol-14-one (19). Yield: 19 mg (79%) obtained as an amorphous powder. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.30 (3H, s), 1.38 (3H, s), 1.39 (3H, d, J = 7.3 Hz), 1.66 (1H, m), 1.91 (3H, m), 2.19 (3H, m), 2.28 (1H, m), 2.78 (1H, d, J = 15.2 Hz), 2.89 (1H, d, J = 15.2 Hz), 3.19 (1H, m), 3.45 (1H, d, J = 10.2 Hz), 5.91 (1H, bs), 7.08 (1H, ddd, J = 1.2, 7.4, 7.4 Hz), 7.15 (1H, ddd, J = 1, 7.8, 7.8 Hz), 7.30 (1H, d, J = 7.8 Hz), 7.40 (1H, d, J = 7.4 Hz), 7.86 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.0, 24.0, 29.9, 30.1, 30.5, 30.6, 34.0, 40.4, 46.3, 53.8, 56.7, 59.8, 65.5, 104.6, 110.6, 117.9, 119.5, 121.8, 126.9, 136.3, 140.8, 173.7. IR (NaCl, neat): 3305, 3060, 2924, 1667, 1455, 1368, 1261, 1109, 1014, 801, 741, 706 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{28}N_3O$: 350.2232. Found 350.2235 (M + H). R_f 0.40 (eluted with 4% MeOH/CH_2Cl_2).

(1*S*,5a*R*,12a*S*,13a*S*)-(±)-*rel*-2,3,11,12,12a,13-Hexahydro-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(iminomethano)-1*H*-indolizino[7,6-*b*]carbazol-14-one (21). Yield: 8.5 mg, (61%); obtained as an amorphous powder. ^{1}H (300 MHz, $CDCl_3$): δ 1.02 (3H, d, J = 7.3 Hz), 1.19 (3H, s), 1.29 (3H, s), 1.42 (1H, m), 1.69 (1H, m), 2.14 (2H, m), 2.22 (1H, m), 2.35 (1H, dd, J = 8.6, 17.2 Hz), 2.67 (1H, d, J = 10.2 Hz), 2.79 (1H, d, J = 17.2 Hz), 2.92 (1H, d, J = 17.2 Hz), 2.96 (1H, m), 3.06 (1H, ddd, J = 2.5, 8.6, 8.6 Hz), 3.14 (1H, d, J = 10.6 Hz), 5.67 (1H, bs), 7.08 (1H, ddd, J = 1.2, 7.4, 7.4 Hz), 7.15 (1H, m), 7.31 (1H, dd, J = 1.1, 7.1 Hz), 7.41 (1H, dd, J = 1.0, 7.7 Hz), 7.87 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$): δ 17.4, 24.8, 27.3, 28.0, 29.2, 30.6, 32.7, 34.3, 45.7, 53.0, 55.2, 62.4, 65.9, 103.2, 110.8, 117.9, 119.6, 121.9, 127.0, 136.3, 141.5, 174.2. IR (NaCl, neat): 3281, 3060, 2960, 1668, 1462, 133, 1123, 1009, 740, 702 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{28}N_3O$: 350.2232. Found 350.2233 (M + H). R_f 0.17 (eluted with 4% MeOH/CH_2Cl_2).

(1*R*,5a*R*,12a*S*,13a*S*)-(±)-*rel*-2,3,11,12,12a,13-Hexahydro-1,12,12-trimethyl-5*H*,6*H*-5a,13a-(iminomethano)-1*H*-indolizino[7,6-*b*]carbazol-14-one (24). Yield: 2.4 mg (54%) obtained as an amorphous powder. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.24 (3H, s), 1.29 (3H, s), 1.41 (3H, d, J = 6.6 Hz), 1.89 (3H, m), 2.07 (1H, dd, J = 4, 13 Hz), 2.07 (1H, m), 2.14 (1H, m), 2.43 (1H, m), 2.57 (1H, d, J = 10.5 Hz), 2.77 (1H, d, J = 17.4 Hz), 2.94 (1H, d, J = 17.4 Hz), 3.19 (1H, d, J = 10.5 Hz), 3.22 (1H, m), 5.48 (1H, bs), 7.09 (1H, ddd, J = 0.8, 7.4, 7.4 Hz), 7.15 (1H, ddd, J = 1.2, 7.6, 7.6 Hz), 7.31 (1H, d, J = 7.8 Hz), 7.40 (1H, d, J = 7.8 Hz), 7.81 (1H, bs). ^{13}C NMR (100 MHz, $CDCl_3$ + 1 drop DMSO-d_6): 11.9, 24.5, 25.3, 27.5, 27.9, 28.8, 29.5, 34.4, 37.8, 43.8, 52.7, 54.6, 59.2, 69.5, 101.5, 110.9, 117.8, 119.1, 121.6, 136.5, 140.6, 176.8. IR (NaCl, neat): 3213, 2959, 1694, 1454, 1259, 1022, 797, 702 cm^{-1}. HRMS (FAB+) Calcd for $C_{22}H_{28}N_3O$: 350.2232. Found 350.2235 (M + H). R_f 0.23 (eluted with 4% MeOH/CH_2Cl_2).

Acknowledgment. This work was supported by the NIH (Grant CA 70375 to RMW) and the DGICYT of Spain (Proyecto PB95-1089 to JFS-C). We also acknowledge the ACS Division of Organic Chemistry Fellowship (sponsored by SmithKline Beecham) and the Pharmacia-Upjohn Company for Fellowship support for Emily M. Stocking. Mass spectra were obtained on instruments supported by the NIH Shared Instrumentation Grant GM49631.

Supporting Information Available: General experimental considerations, tables of complete ^{1}H NMR NOE data for cycloadducts **12**−**15**, and ^{1}H NMR spectra of natural and synthetic VM55599. This material is available free of charge via the Internet at http://pubs.acs.org.

JA993791H