Synthesis of Structurally Simplified Analogues of Pancratistatin: Truncation of the Cyclitol Ring

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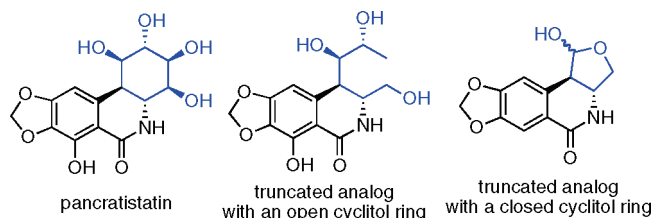
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pancratistatin

truncated analog with an open cyclitol ring

truncated analog with a closed cyclitol ring

Pancratistatin is a phenanthridone-type natural product isolated from several plants of the Amaryllidaceae family. Its potent antiproliferative, antivascular, antiviral, and antiparasitic properties have attracted the attention of synthetic, biological, and medicinal chemists. Pancratistatin's low natural availability and complex structure have steered many of these research projects toward the preparation of its simplified synthetic analogues with useful levels of activity. In this work we have developed synthetic chemistry aimed at the preparation of pancratistatin analogues with a truncated cyclitol portion of the molecule. The described synthetic pathways are based on a highly *anti*-diastereoselective arylcuprate conjugate addition to γ-alkoxy-α,β-enoates and *syn*-selective azidation at the α-position of ester enolates. Analogues with the formally cleaved C3−C4 bond, and thus containing an open ring C, as well as a compound containing a truncated lactol moiety in lieu of the cyclitol, were prepared. Several of the analogues exhibited weak antiproliferative activity, with the highest potency observed in the case of the lactol analogue. From these results implications for the design of future pancratistatin analogues are discussed. Furthermore, the synthetic pathways can be used to construct pancratistatin-mimetic libraries, in which the cyclitol moiety is replaced by other cyclic motifs.

Introduction

The medicinal value of Amaryllidaceae plant extracts has been recognized for a long time. It dates back to at least the 4th century BC, when Hippocrates of Cos used oil from the daffodil *Narcissus poetica L.* for the treatment of cancer.[1] In more recent times, more than 100 structurally diverse alkaloids, possessing a wide spectrum of biological activities have been isolated from Amaryllidaceae species.[2] Lycorine

(Figure 1), shown to possess antitumor and antiviral activity,[3] was the first member of this family isolated in 1877.[4] Later, the phenanthridone narciclasine attracted considerable interest due to its antineoplastic properties[5] and most recently due to its unique mode of action in glioblastoma

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cells and high potential in treating various forms of brain cancer.[6]

Structurally similar phenanthridones (+)-pancratistatin, isolated by Pettit and co-workers,[7] and (+)-7-deoxypancratistatin (Figure 1), isolated by Ghosal and co-workers[8] have also received considerable attention from both synthetic[9] and medicinal chemistry communities.[10] Pancratistatin has been found to exhibit strong *in vitro* antiproliferative effect against the U.S. National Cancer Institute (NCI) panel of cancer cell lines as well as a number of *in vivo* experimental cancer systems.[11] A number of recent reports demonstrate that pancratistatin is specifically toxic to cancer cells as opposed to normal ones, whereas the currently used anticancer drugs, such as taxol and etoposide, are equally toxic to both cell types.[12] Powerful antiviral[13] and
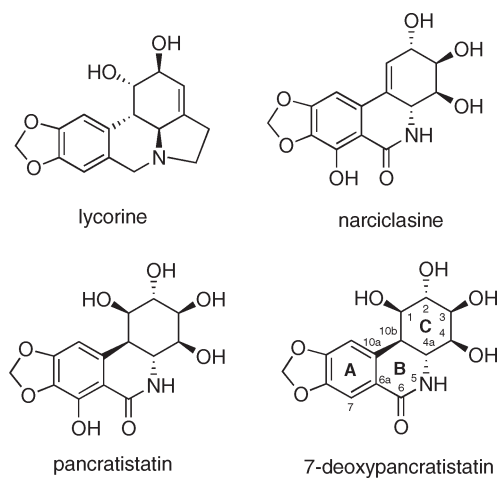

FIGURE 1. Structures of selected Amaryllidaceae constituents with promising anticancer activities.

antiparasitic[14] activities of pancratistatin constitute a related area of promise. Importantly, it has been noted that pancratistatin and 7-deoxypancratistatin are the only known agents (other than an interferon inducer) to show a significant chemotherapeutic efficacy in a Japanese encephalitis virus-infected mouse model.[13] Although less potent, (+)-7-deoxypancratistatin exhibits a better therapeutic index in *in vitro* antiviral (RNA) assays due to reduced toxicity.[13] Unfortunately, studies directed at elucidation of pancratistatin's mode of action have been hampered by its low natural abundance (ca. 0.039% of dry *Pancratium littorale* bulbs) and therefore the extremely small quantity of material available from isolation.[7] This lack of supply, as well as poor water solubility (< 53 μg/mL), has also plagued the clinical development of pancratistatin either as an anticancer or an antiviral agent. Although the water solubility problem has been addressed by developing phosphate prodrugs, this was accomplished utilizing syntheses from the already poorly available natural product.[10o,p]

In view of the limited supply and the promising activities associated with these natural products, extensive synthetic studies have been undertaken, and several total syntheses of both pancratistatin and its 7-deoxy congener have been reported. An overview of this body of work suggests that these molecules represent formidable synthetic challenges and that they are difficult to construct in a short practical way.[9] Therefore, research programs directed at preparation of synthetic analogues with simplified structures have been pursued by a number of laboratories. So far such efforts have not resulted in identification of truncated pancratistatin analogues with comparable levels of biological activities. Thus, Hudlicky and co-workers reported that the reduction in oxygenation of ring A or its bioisosteric substitution with an indole moiety led to a significant drop in potency.[10c−e] Ring B opening, either by cleaving the C10a-C10b bond or removing the amide functionality, also resulted in inactive analogues as reported by Chapleur's and our group, respectively.[10i,j] *seco*-B,C-Pancratistatin

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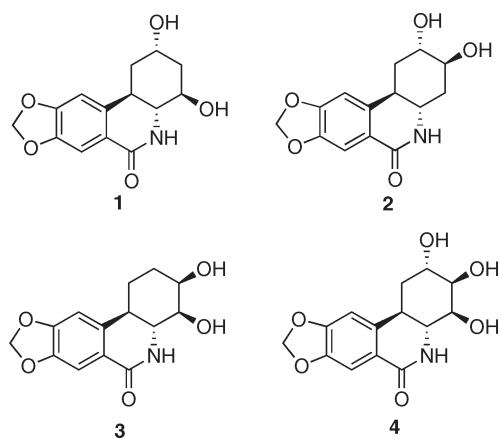
FIGURE 2. Selected dehydroxylated pancratistatin analogues.

analogues, synthesized by McNulty and co-workers and containing opened B and C rings, were shown to be inactive.[10b] In addition, systematic removal of hydroxyl groups in ring C leads to compounds with significantly diminished activity as well. Thus, analogues containing two hydroxyl groups in the cyclitol portion of the molecule were prepared and biologically evaluated by McNulty and Hudlicky's groups, respectively (Figure 2).[10c,k,l]

Compound **1** was found to be inactive, **2** was on average 2−3 orders of magnitude less potent than pancratistatin in the NCI 60 cell line screen, and **3** showed only low micro-molar growth-inhibitory potency against the murine P388 cell line but was completely inactive against a panel of human cancer cell lines. To date, 7-deoxy-*trans*-dihydro-narciclasine (**4**), possessing three hydroxyl groups in ring C, represents the only ring C dehydroxylated analogue with significant levels of activity.[11b,15] Therefore, we wondered whether structural simplification of pancratistatin could be achieved not by removing the hydroxyl functionality, as was attempted with the above-mentioned efforts, but by modification of the carbon skeleton of the ring C while still preserving at least three hydroxyl groups. We set out to develop a synthetic pathway to ring C-opened analogues of pancratistatin and 7-deoxypancratistatin shown in Figure 3 (**5** and **6**). In addition, we considered a possibility of cyclizing either compound **5** or **6** into a lactol (e.g., **7**), which would have a considerably truncated, but closed, ring C moiety. In this article, we describe the results of this endeavor and the biological evaluation of the first pancratistatin analogues with the truncated cyclitol portion of the molecule.

Results and Discussion

Because of the successful application of the Banwell variant of the Bischler−Napierlaski cyclization in this area of research,[9,16] we considered the preparation of compounds **5** and **6** from the required carbamates **A**, which in turn could

FIGURE 3. Proposed pancratistatin analogues with a truncated cyclitol ring.
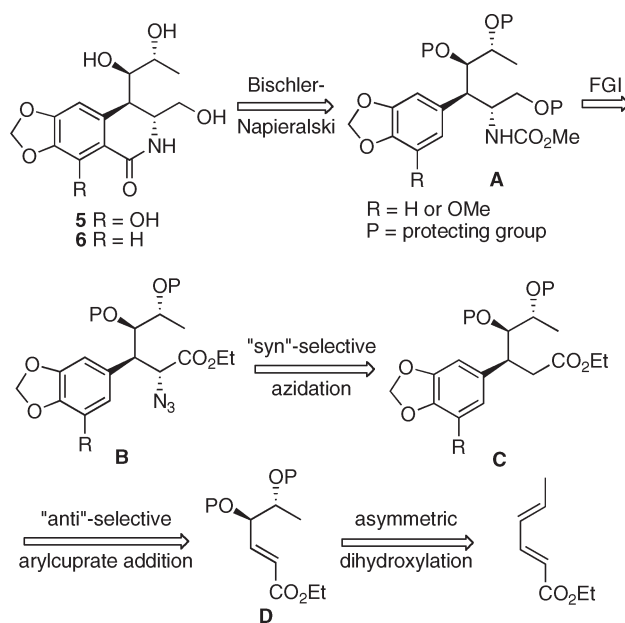
FIGURE 4. Retrosynthetic analysis of pancratistatin analogues with an open cyclitol ring.

be obtained from azides **B** by way of an appropriate functional group interconversion (FGI, Figure 4). Significant literature precedent for *syn*-selective introduction of nitrogen functionality (including azidation)[17] at the α-position of esters boded well for the conversion of esters **C** to azido esters **B** with the required stereochemistry. Previously, we have studied arylcuprate addition with γ-alkoxy-α,β-unsaturated esters in detail and derived a reductive-elimination-based model to predict the stereochemical outcome in such reactions.[18] Therefore, we had a high degree of optimism that we

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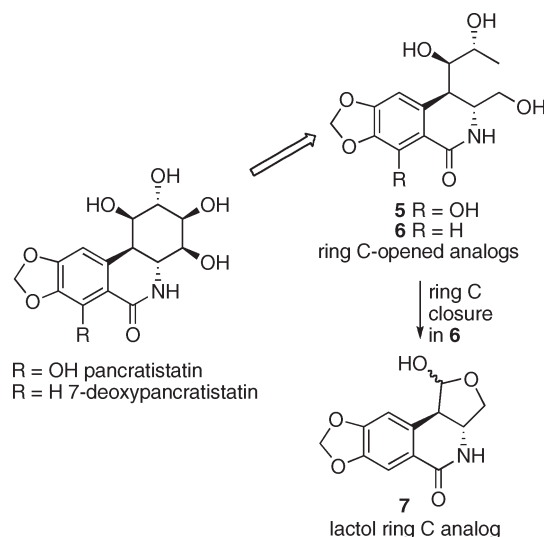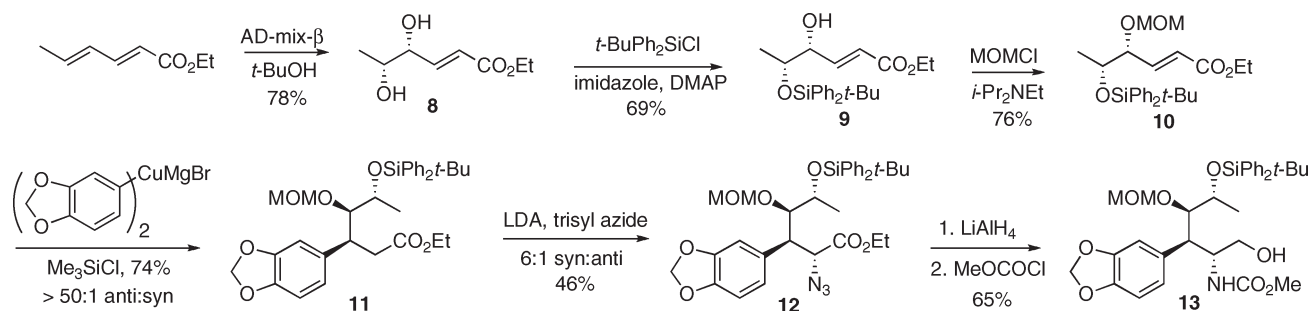
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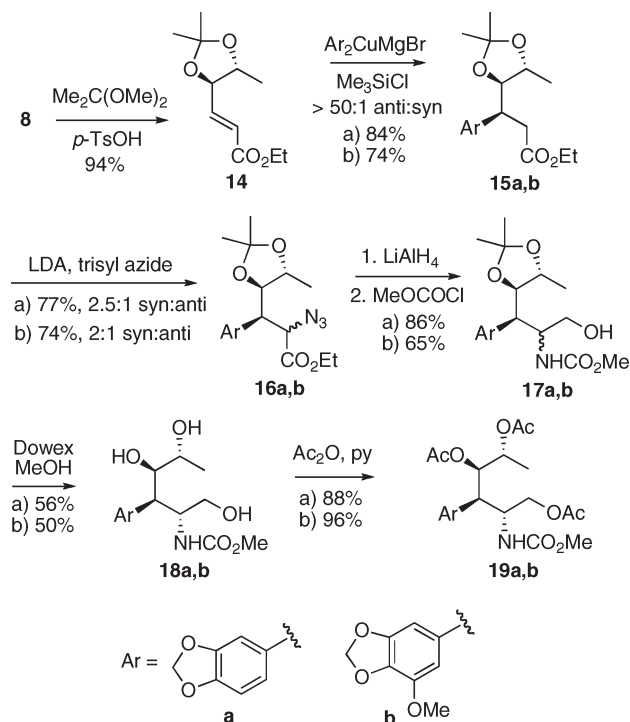
SCHEME 1.   Synthesis of Carbamate 13

would be able to accomplish the highly *anti*-selective transformation **D → C**, utilizing the required arylcuprates. Finally, asymmetric dihydroxylation of commercially available inexpensive sorbate esters has been previously reported by Sharpless and co-workers.[19]

Indeed, dihydroxyenoate **8** was obtained as a single *R,R*-enantiomer when ethyl sorbate was reacted with AD-mix-β as was established by comparing its optical rotation with the literature value (Scheme 1).[19]

Selective protection of the distant hydroxyl as a TBDPS ether, followed by the treatment of the resulting alcohol **9** with MOMCl, gave enoate **10** in good yield. The arylcuprate addition of a Grignard-derived organocopper reagent in the presence of TMSCl gave ester **11** as the exclusive *anti*-diastereomer. This highly favorable stereochemical outcome was expected on the basis of the previous systematic studies of this process in our laboratory. *syn*-Selective azidation of **11** was achieved by treatment with KHMDS and trisyl azide, following the precedent of Hanessian and co-workers.[17] However, the yields and selectivities were not reproducible and depended on the source of the KHMDS reagent. Therefore, we employed freshly prepared LDA (BuLi + *i*-Pr$_2$NH) as a base. This gave us somewhat lower *syn*-selectivity (6:1) but allowed for a facile scale-up and was highly reproducible. Simultaneous reduction of the ester and azide functionalities in **12** with LiAlH$_4$ followed by treatment of the crude product with methyl chloroformate resulted in carbamate **13**. In preparation for the Bischler–Napieralski cyclization to form the lactam ring B, we attempted to replace the acid-sensitive protecting groups in carbamate **13** with acetates. Unexpectedly, all efforts to remove the bulky TBDPS group resulted in mixtures of products that appeared to contain no methoxy-carbamate as was established by NMR analysis. While this rather curious setback did not seem insurmountable, we needed to prepare more material by scaling up the synthesis from diol **8**. In view of the difficulty of the deprotection of the silyl ether in carbamate **13**, we chose not to repeat the same reaction sequence but rather to modify our protecting group strategy. We thus masked the two hydroxyl groups in diol **8** with an isopropylidene functionality (Scheme 2) to obtain compound **14**.

Despite significant conformational constraints, enoate **14** underwent the organocopper addition reactions with cuprates derived from **a** and **b** with exclusive *anti*-selectivity. The *syn*-selectivity in the azidation of the enolates derived from

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SCHEME 2.   Synthesis of Bischler–Napieralski Precursors 19a and 19b

15a and **15b** was, however, somewhat lower than in the corresponding transformation **11 → 12**. Reduction of the azide and ester functionalities in the epimeric mixtures **16a** and **16b** was followed by their conversion to epimeric mixtures of carbamates **17a** and **17b** as described before. In each of these mixtures the desired major epimer was purified from the minor one after isopropylidene deprotection, to give pure triols **18a** and **18b**. These were then converted into their corresponding triacetates **19a** and **19b** in preparation for the Bischler–Napieralski cyclization. While the unambiguous stereochemical assignment of the reaction intermediates would be obtained later (X-ray structure of **23**, vide infra), at this stage the identities of the major and minor epimers in **16a** were confirmed by conversion to the corresponding lactones **20** and **21** (Figure 5) and NMR analysis of these conformationally constrained cyclic systems. The large (J = 12 Hz) coupling constant between H$_\alpha$ and H$_\beta$ in the major lactone **20** and the smaller one (J = 8 Hz) in the minor one indicated the corresponding *trans* and *cis* positioning of H$_\alpha$ and H$_\beta$ in **20** and **21**, in accordance with the assigned stereochemistry.
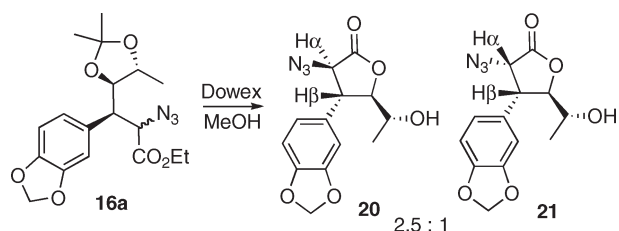
FIGURE 5. Confirmation of azidation stereochemistry.

The commonly utilized Banwell conditions for the Bischler−Napieralski condensation[9,16] provided only modest yields of the cyclized products when either **19a** or **19b** was subjected to the treatment with Tf₂O and DMAP in CH₂Cl₂ (Scheme 3). After many unsuccessful optimization attempts we opted to test whether the recently described use of 2-chloropyridine as a base, in a related cyclization of amides to dihydroisoquinolines,[20] would work on our carbamate substrates. Gratifyingly, the reaction yields were raised to respectable levels without a change in regioselectivity. Imidate **22** was the only isolated regioisomer in the cyclization of **19a**, while the preponderance of the regioisomer resulting from the attack *para* to the methylenedioxy functionality was obtained in the cyclization of **19b**. The regiochemistries of **22**, **24**, and **25** were established by ^{1}H NMR analysis of the aromatic portions of the molecules, and both of these regiochemical outcomes are consistent with the literature precedent on related systems.[9c] The imidates were smoothly converted to the corresponding amides with either acidic hydrolysis or treatment with Me₃SiI. Notably, the phenolic methyl ether was concomitantly removed to give directly triacetate **27** when Me₃SiI was utilized. Alternatively, **27** was also obtained under these conditions from the product of hydrolysis, lactam **26**. The acetates were subsequently removed in both lactams **23** and **27** to give the desired pancratistatin truncated analogues **5** and **6** with open cyclitol rings. In addition, the vicinal diol functionality in **6** was smoothly cleaved with NaIO₄ to give the corresponding aldehyde that closed to the single anomeric lactol form **7** in DMSO-d_6, as was established by ^{1}H NMR analysis.

Analogues **5** and **6**, their triacetates **23** and **27**, and lactol **7** were evaluated for antiproliferative activity utilizing HeLa and MCF-7/AZ (models for cervical and breast adenocarcinomas) human cancer cell lines. Because of the unavailability of pancratistatin, the biological evaluation was performed with the use of phenathridone narciclasine as the positive control (see Experimental Section for its isolation from a plant source). This choice is justified by the similar activity profiles and potencies of narciclasine and pancratistatin.[11] In contrast to narciclasine, which exhibited potent cell killing properties, the synthetic compounds manifested useful activity only at high micromolar concentrations. At a concentration of 300 μM compound **5**, containing the phenolic hydroxyl, reduced the viability of HeLa and MCF7/AZ cells to 70% and 82% respectively (Table 1). Its triacetate **27** exhibited slightly enhanced activity (60% and 64%). It is likely that masking the three hydroxyl groups in **5** improves cell penetration, while the acetates undergo rapid hydrolytic removal within cells. Indeed, under the same testing conditions narciclasine tetraacetate displayed

antiproliferative activity that was similar in magnitude to that of narciclasine, as we reported previously.[21] Trihydroxy analogue **6** or its triacetate were virtually inactive. However, transformation of the ring C-opened analogue **6** into lactol **7** gave the highest potency (59% and 48% cell viability) among the synthesized compounds.

While the antiproliferative potencies are rather low, we believe that these results are instructive. The lactam ring B in pancratistatin is strained because of the sp₂-character of the four atoms at positions N5, C6, C6a and C10a, which are forced out of planarity by the *trans* B−C ring fusion. This strain is relieved upon ring C opening, leading to a significant change in the overall geometry. The X-ray structure of triacetate **23** (Figure 6a) illustrates this point. The planarity requirement in ring B forces the C1−C11 and C2−C18 bonds to adopt a quasidiaxial orientation with a torsion angle of 170° for the bond C11−C1−C2−C18. Using the crystallographic structure of **23** and the published X-ray structure of pancratistatin[22] for initial geometries, we performed DFT conformational optimizations of ring C-open analogue **6**, pancratistatin and ring C-closed analogue **7** (Figure 6b−d). It can be readily seen that the planar molecular framework of pancratistatin (c) is drastically distorted in analogue **6** (b) but not in **7** (d). Thus, the crystallographic and computational data provide a likely explanation for a virtually complete loss of activity upon ring C opening in pancratistatin and retention of a small fraction of antiproliferative effect in a significantly truncated lactol ring C analogue.

The results of the current investigation represent a contribution to the previously reported structure−activity relationship data associated with the C-ring of pancratistatin.[10b,c,k,l] Thus, it appears that not only are the number and positioning of the hydroxyl groups important for the potent anticancer properties of pancratistatin analogues but also the cyclic nature of the cyclitol portion of the molecule is a key component of the natural product's cytotoxic pharmacophore. The opening of the cyclitol ring increases conformational flexibility and, perhaps more importantly, results in a drastic conformational change that distorts the overall planarity of the carbon framework. It is becoming increasingly apparent that the only structural changes of the ring C that lead to potent compounds are modifications at the C1 and C10b positions (Figure 1). This includes the C1−C10b double bond, found in narciclasine and 7-deoxynarciclasine (known as lycoricidine),[5b] as well as the removal of the C1 hydroxyl, as found in *trans*-dihydronarciclasine and 7-deoxy-*trans*-dihydronarciclasine (**4**, Figure 2). All these compounds possess nanomolar antiproliferative activities toward human cancer cells.[5b,11b,15] Furthermore, Pettit and co-workers showed that the antiproliferative potency of C1-benzoate ester of pancratistatin exceeds that of pancratistatin by 10−100 times depending on the cell type.[23] However, it is not clear whether the benzoate moiety is an important part of the cytotoxic pharmacophore or enhances anticancer

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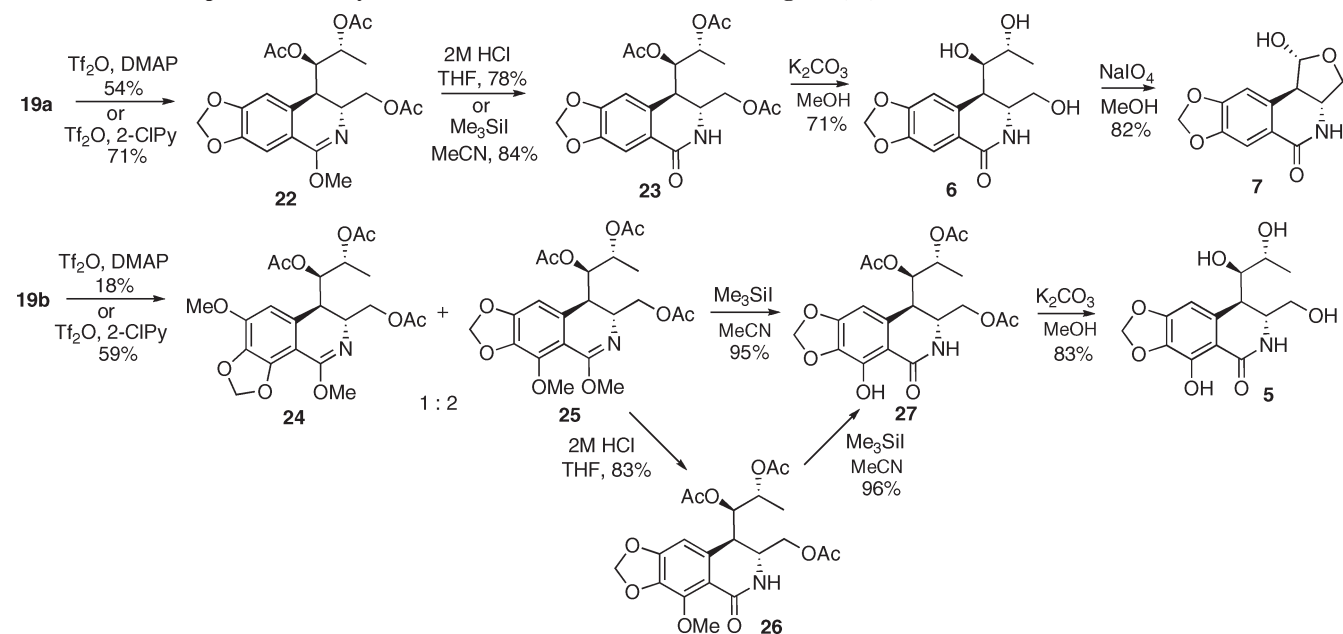**SCHEME 3.** Completion of the Synthesis of Truncated Pancratistatin Analogues 5, 6, and 7

TABLE 1. Antiproliferative Activity of 5, 6, 7, 23, and 27

	% cell viability[a]	
compound	HeLa	MCF-7/AZ
6	78 ± 3	83 ± 5
5	70 ± 9	82 ± 11
7	59 ± 8	48 ± 3
23	89 ± 4	92 ± 3
27	60 ± 3	64 ± 6
narciclasine	6 ± 1	8 ± 2

[a]% cell viability after 48 h of treatment with indicated compounds at 300 μM relative to DMSO control ± SD from two independent experiments, each performed in 8 replicates, determined by MTT assay.
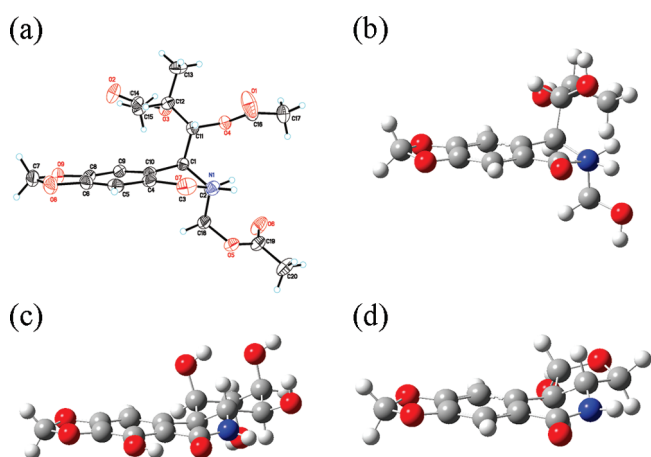

FIGURE 6. (a) X-ray structure of analogue **23** (thermal ellipsoids are shown at 25% probability). DFT optimizations of molecular structures of (b) analogue **6**, (c) pancratistatin, and (d) analogue **7**.

activity by facilitating the intracellular delivery followed by hydrolysis within cells.

Because of a significant conformational change brought about by the C-ring opening, it is possible that the observed low levels of activity found with the C-ring truncated compounds synthesized in this work arise from an independent

mode of action. However, a change in mechanism is a possibility for all truncated pancratistatin analogues, whose structures are significantly different from that of the natural product. The solution to this potential obstacle will be achieved after the biological mechanism(s) responsible for the anticancer effect of pancratistatin are unravelled, making it possible to evaluate its analogues in a direct mechanism-based assay.

Conclusions

As a result of the significant medicinal potential and low natural abundance of the Amaryllidaceae constituent pancratistatin, there is a great demand for synthetic analogues with drastically simplified structures that can be rapidly accessed from commercially available inexpensive starting materials. Synthetic chemistry described in this work leads to a facile preparation of pancratistatin analogues with the truncated cyclitol portion of the molecule. The results of the biological evaluation of these compounds, complemented with DFT computations, suggest that the polyhydroxylated portion of the molecule must remain cyclic for antiproliferative activity to be observed. This also preserves the overall planarity of the carbon framework, which is evidently critical. Since the cellular macromolecular target for pancratistatin is yet to be identified, this study provides further guidance in the quest for structurally simplified potent pancratistatin analogues. Compounds **5** and **6** could thus be used as starting points for the construction of pancratistatin-mimetic libraries, incorporating diverse cyclic moieties in lieu of the six-membered cyclitol ring. The fact that the replacement of the stereochemically complex cyclitol part of the molecule with a simple five-membered lactol results in retention of small levels of activity bodes well for these future endeavors.

Experimental Section

Isolation of Narciclasine. Fresh bulbs (7.5 kg) of *Narcissus pseudonarcissus* (var. King Alfred) were dried at 40 °C for 3−4 days.

The dried material (3 kg) was milled and extracted with 0.1 N NaOH according to the previously reported procedure.[24] The combined basic extracts were acidified to pH 2 with 96% H_2SO_4 and extracted with EtOAc (3 × 15 L). The combined organic extracts were dried (Na_2SO_4) and evaporated under reduced pressure. The residue (61 g) was separated in aliquots of 15 g and fractionated by silica gel column (CHCl$_3$/i-PrOH 7:3 v/v). The column fractions that by TLC analysis contained narciclasine were combined in two groups. These were further purified by preparative TLC, using CHCl$_3$/i-PrOH 7:3 or CHCl$_3$/MeOH 9:1 v/v affording pure narciclasine as amorphous solid (513 mg, 142.5 mg/kg).

(4R,5R,E)-Ethyl 5-(tert-Butyldiphenylsilyloxy)-4-hydroxy-hex-2-enoate (9). Diol **8**[19] (1 g, 5.7 mmol) was dissolved in DMF (7 mL) and cooled to 0 °C. To the mixture were added DMAP (7 mg, 0.57 mmol), imidazole (0.43 g, 6.31 mmol), and tert-butyl(chloro)diphenylsilane (1.7 g, 6.31 mmol). The resulting mixture was stirred overnight at room temperature. The reaction mixture was quenched with water (10 mL). The aqueous phase was extracted with CH_2Cl_2 (3 × 15 mL). The combined organic layers were washed with brine, dried (MgSO$_4$), and evaporated under reduced pressure. The residual oil was presorbed on silica gel and purified by column chromatography (5−15% EtOAc/hexanes) to afford compound **9** in 69% yield as a colorless oil; R_f 0.58 (20% EtOAc/hexanes); ^{1}H NMR (CDCl$_3$) δ 7.64−7.71 (m, 4 H), 7.37−7.44 (m, 6H), 6.90 (dd, J = 15.7, 4.4 Hz, 1H), 6.12 (dd, J = 15.7, 1.7 Hz, 1H), 4.20 (q, J = 7.0 Hz, 2H), 4.12 (m, 1H), 3.82 (m, 1H), 2.59 (d, OH), 1.28 (t, J = 7.0 Hz, 3H), 1.05 (s, 12H); ^{13}C NMR (CDCl$_3$) δ 166.3, 147.1, 135.9, 130.0, 129.7, 127.7, 127.7, 122.2, 75.4, 72.4, 60.4, 53.5, 27.0, 19.8, 19.4, 14.3; HRMS m/z (ESI) calcd for $C_{24}H_{32}O_4SiNa$ (M + Na)$^+$ 435.1964, found 435.1968.

(4R,5R,E)-Ethyl 5-(tert-Butyldiphenylsilyloxy)-4-(methoxymethoxy)hex-2-enoate (10). To a solution of **9** (1.3 g, 3.15 mmol) in CH_2Cl_2 (60 mL) at 0 °C was added i-Pr$_2$NEt (4 g, 31.5 mmol) followed by the dropwise addition of chloro(methoxy)methane (1.27 g, 15.8 mmol). The resulting solution was stirred for 20 h while it was allowed to warm to room temperature. The reaction mixture was quenched with satd NH$_4$Cl (30 mL). The aqueous phase was extracted with ether (3 × 50 mL). The combined organic layers were washed with brine, dried (MgSO$_4$) and evaporated to afford dark yellow oil. The crude material was presorbed on silica gel and purified by column chromatography (5−15% EtOAc/hexanes) to afford enoate **10** in 76% yield as a colorless oil; R_f 0.65 (20% EtOAc/hexanes); ^{1}H NMR (CDCl$_3$) δ 7.34−7.67 (m, 10H), 7.06 (dd, J = 15.7, 5.0 Hz, 1H), 6.06 (dd, J = 15.7, 1.7 Hz, 1H), 4.45 (d, J = 10.6 Hz, 1H), 4.39 (d, J = 10.6 Hz, 1H), 4.21 (q, J = 7.2 Hz, 2H), 4.16 (m, 1H), 3.96 (m, 1H), 3.18 (s, 3H), 3.30 (t, J = 7.2 Hz, 3H), 1.07 (s, 9H), 1.01 (d, J = 6.3 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 145.2, 136.3, 131.0, 128.1, 127.6, 123.1, 95.4, 78.6, 70.5, 60.5, 55.7, 27.1, 18.1, 14.3; HRMS m/z (ESI) calcd for $C_{26}H_{36}O_5SiNa$ (M + Na)$^+$ 479.2230, found 479.2230.

General Procedure for Arylcuprate Addition. A few drops of the corresponding aryl bromide saturated solution in THF were added to crushed Mg turnings (7.03 mmol) in THF (10 mL) under nitrogen atmosphere. Once the reaction started the solution warmed up and slightly darkened. The rest of the aryl bromide (7.03 mmol total) was added dropwise to allow a gentle reaction. The reaction mixture was allowed to cool to room temperature and then cannulated to a slurry of CuI (3.52 mmol) in THF (10 mL) at −78 °C. The mixture was stirred at −78 °C for 40 min. Me$_3$SiCl (7.03 mmol) and the enoate (0.703 mmol in 10 mL of THF) were added sequentially dropwise at −78 °C. The yellow-brown suspension was stirred overnight while slowly warming up to room temperature. The reaction mixture was

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quenched with a mixture of concd NH$_4$OH and satd NH$_4$Cl (1: 9, 30 mL) and extracted with ether (3 × 30 mL). The combined organic layers were washed with brine, dried with MgSO$_4$, and concentrated under reduced pressure. The residue was absorbed on silica gel and purified by column chromatography (5−30% EtOAc/hexanes) to yield corresponding addition product as an oil.

(3S,4R,5R)-Ethyl-3-(benzo[d][1,3]dioxol-5-yl)-5-(tert-butyldiphenylsilyloxy)-4-(methoxymethoxy)hexanoate (11). Yield 74%, R_f 0.48 (20% EtOAc/hexanes); ^{1}H NMR (CDCl$_3$) δ 7.58 (d, J = 6.9 Hz, 2H), 7.38 (d, J = 6.9 Hz, 2H), 7.34 (m, 6H), 6.66 (m, 3H), 5.87 (d, J = 10.5 Hz, 2H), 4.59 (d, J = 6.9 Hz, 1H), 4.52 (d, J = 6.9 Hz, 1H), 4.00 (q, J = 7.2 Hz, 2H), 3.89 (m, 1H), 2.88 (dd, J = 14.9, 3.85 Hz, 1H), 2.54 (dd, J = 14.9, 9.0 Hz, 1H), 1.12 (t, J = 7.2 Hz, 3H), 1.01 (d, J = 6.9 Hz, 3 H), 0.98 (s,9H); ^{13}C NMR (CDCl$_3$) δ 172.7, 147.6, 146.1, 136.6, 136.0, 135.9, 134.6, 133.8, 129.6, 127.6, 127.4, 121.7, 108.7, 108.1, 100.8, 98.2, 85.5, 70.4, 60.2, 56.1, 42.3, 38.6, 27.1, 19.2, 14.2.; HRMS m/z (ESI) calcd for $C_{33}H_{42}O_7SiNa$ (M + Na)$^+$ 601.2595, found 601.2598.

Ethyl (3S)-3-(1,3-Benzodioxol-5-yl)-3-[(4R,5R)-2,2,5-trimethyl-1,3-dioxolan-4-yl]propanoate (15a). Yield 84%, R_f 0.61 (20% EtOAc/hexanes); [α]$^{22}_D$ +40.2 (c 0.02, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.63−6.75 (m, 3H), 5.92 (s, 2H), 4.05 (q, J = 7.2 Hz, 2H)), 3.78 (m, 1H), 3.56 (t, J = 8.3 Hz, 1H), 3.13 (m, 1H), 2.98 (dd, J = 15.7, 5.0 Hz, 1H), 2.58 (dd, J = 15.4, 10.2 Hz, 1H), 1.30 (s, 6H), 1.12 (t, J = 7.2 Hz, 3H), 0.76 (d, J = 6.1 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 172.2, 147.8, 146.7, 133.7, 121.5, 108.42, 108.4, 101.1, 85.4, 60.3, 46.0, 38.6, 27.5, 27.1, 18.8, 14.2; HRMS m/z (ESI) calcd for $C_{18}H_{24}O_6Na$ (M + Na)$^+$ 359.1471, found 359.1471.

Ethyl (3S)-3-(7-Methoxy-1,3-benzodioxol-5-yl)-3-[(4R,5R)-2,2,5-trimethyl-1,3-dio-xolan-4-yl]propanoate (15b). Yield 74%; R_f 0.21 (20% EtOAc/hexanes); [α]$^{22}_D$ +34.8 (c 0.09, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.40 (d, J = 1.4 Hz, 1H), 6.36 (d, J = 1.1 Hz, 1H), 5.92 (s, 2H), 4.11−3.75 (m, 3H), 3.87 (s, 3H), 3.57 (t, J = 8.3 Hz, 1H), 3.09 (m, 1H), 2.96 (dd, J = 15.7, 4.9 Hz, 1H), 2.56 (q, J = 9.9 Hz, 1H), 1.37 (s, 6H), 1.13 (t, J = 7.43 Hz, 3H), 0.78 (d, J = 6.0 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 172.2, 149.0, 143.5, 134.5, 108.0, 102.0, 101.5, 85.3, 60.4, 56.7, 46.3, 38.6, 27.4, 27.0, 18.7, 14.1; HRMS m/z (ESI) calcd for $C_{19}H_{26}O_7Na$ (M + Na)$^+$ 389.1576, found 389.1583.

General Procedure for Azidation with LDA. To a solution of freshly distilled diisopropylamine (8.19 mmol) in 10 mL dry THF at −78 °C was added 7.93 mmol of n-BuLi dropwise under N$_2$ atmosphere. After stirring the solution at this temperature for 45 min, 1.3 mmol of the respective arylesters in THF (7 mL) was added slowly, and the resulting solution was warmed to −30 °C and stirred for 2 h. The solution was cooled back to −78 °C, and HMPA (20.8 mmol) was added in one portion followed by the dropwise addition of precooled trisylazide (5.2 mmol). The resulting mixture was stirred at −78 °C for 2 h, and then glacial acetic acid (19 mmol) was added. The reaction mixture was allowed to warm to room temperature and to stir at room temperature for 12 h. Then 50 mL of satd NaHCO$_3$ was added and extracted with (3 × 50 mL) CH_2Cl_2. The combined organic layers were washed with brine (1 × 30 mL) and water (2 × 30 mL), dried (MgSO$_4$), and evaporated under reduced pressure. The residual oil was presorbed on silica gel and purified by column chromatography (2−5% EtOAc/hexanes) to yield a corresponding azide addition product.

(2R,3R,4R,5R)-Ethyl-2-azido-3-(benzo[d][1,3]dioxol-5-yl)-5-(tert-butyldiphenylsilyloxy)-4-(methoxymethoxy)hexanoate (12). Yield 46%, R_f 0.49 (20% EtOAc/hexanes); ^{1}H NMR (CDCl$_3$) δ 7.23−7.58 (m, 10H), 6.62 (m, 3H), 5.86 (d, J = 10.5 Hz, 2H), 4.80 (d, J = 6.4 Hz, 1H), 4.74 (m, 2H), 4.13 (q, J = 7.2 Hz, 2H), 3.72 (m, 3H), 3.43 (s, 3H), 1.23 (t, J = 7.2 Hz, 3H), 0.96 (s, 12H); ^{13}C NMR (CDCl$_3$) δ 169.9, 147.4, 146.9, 136.0, 135.8, 135.8, 133.9, 129.8, 129.7, 129.6, 127.5, 127.3, 123.5, 110.2, 107.9, 101.0, 98.9, 83.4, 69.6,

64.0, 61.7, 56.4, 47.8, 27.1, 20.0, 19.3, 14.3; HRMS *m/z* (ESI) calcd for $C_{33}H_{41}N_3O_7SiNa$ (M + Na)$^+$ 642.2610, found 642.2611.

Ethyl (2R+S,3R)-2-Azido-3-(1,3-benzodioxol-5-yl)-3-[(4R,5R)-2,2,5-trimethyl-1,3-dioxolan-4-yl]propanoate (16a). Yield 77% (*anti:syn* = 1:2.5); R_f 0.61 (20% EtOAc/hexanes); $[\alpha]^{22}_D$ +27.86 (*c* 0.05, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.67−6.89 (m, 3H), 5.94 (m, 2H), 4.69 (d, *J* = 4.9 Hz, 1H), 4.07−4.29 (m, 3H), 3.91 (m, 1H), 3.77 (m, 1H), 3.31 (dd, *J* = 9.6, 3.6 Hz, 1H), 3.17 (dd, *J* = 9.9, 4.6 Hz, 1H), 1.42 (m, 6H), 1.29 (t, *J* = 7.2 Hz, 3H), 1.18 (t, *J* = 7.2 Hz, 3H), 0.77 (d, *J* = 6.1 Hz, 3H), 0.72 (d, *J* = 6.1 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 169.4, 169.3, 148.1, 147.7, 147.5, 131.1, 128.5, 123.0, 122.4, 109.6, 109.1, 108.5, 108.3, 108.2, 101.3, 101.2, 81.6, 65.6, 64.3, 61.9, 61.8, 51.3, 27.5, 27.2, 26.8, 19.1, 19.0, 14.2; HRMS *m/z* (ESI) calcd for $C_{18}H_{23}N_3O_6Na$ (M + Na)$^+$ 400.1486, found 400.1485.

Ethyl (2R+S,3R)-2-Azido-3-(7-methoxy-1,3-benzodioxol-5-yl)-3-[(4R,5R)-2,2,5-trimethyl-1,3-dioxolan-4-yl]propanoate (16b). Yield 74% (*anti:syn* = 1:1); R_f 0.28 (20% EtOAc/hexanes); selected ^{1}H NMR (CDCl$_3$) data for the *anti*-isomer δ 6.52 (d, *J* = 1.6 Hz, 1H), 6.40 (d, *J* = 0.8 Hz, 1H), 5.92 (s, 2H), 4.63 (d, *J* = 5.2 Hz, 1H), 4.11−3.72 (m, 4H), 3.84 (s, 3H), 3.10 (dd, *J* = 10.5, 5.2 Hz, 1H), 1.39 (s, 3H), 1.38 (s, 3H), 1.15 (t, *J* = 6.9 Hz, 3H), 0.72 (d, *J* = 5.8 Hz, 3H); selected ^{1}H NMR (CDCl$_3$) data for the *syn*-isomer δ 6.46 (m, 2H), 5.90 (m, 2H), 4.28−3.91 (m, 5H), 3.87 (s, 3H), 3.26 (dd, *J* = 9.9, 3.6 Hz, 1H), 1.36 (s, 3H), 1.32 (s, 3H), 1.26 (t, *J* = 7.7 Hz, 3H), 0.76 (d, *J* = 6.0 Hz, 3H); selected ^{13}C NMR (CDCl$_3$) δ 169.4, 149.1, 143.6, 135.1, 131.5, 129.2, 109.3, 108.4, 103.5, 102.9, 81.6, 65.6, 64.3, 61.9, 56.8, 51.5, 27.4, 19.3, 14.3; HRMS *m/z* (ESI) calcd for $C_{19}H_{25}N_3O_7Na$ (M + Na)$^+$ 430.1590, found 430.1587.

General Procedure for Carbamate Formation. To a suspension of LiAlH$_4$ (2.5 mmol) in anhydrous ether (20 mL) at 0 °C was added a solution of the azide (1 mmol in 5 mL) dropwise. The reaction mixture was stirred for 2.5 h at room temperature. After the completion of the reaction, monitored by TLC, 20 mL of ether was added to the reaction mixture and recooled to 0 °C. Saturated NaSO$_4$ (15 mL) was added slowly, and the reaction was stirred for 30 min at room temperature. Fifteen milliliters of water was added and extracted with EtOAc (3 × 25 mL). Combined organic layers were washed with brine, dried (MgSO$_4$), and evaporated under reduced pressure to yield the respective aminoalcohol, which was used without further purification. To the amine (1.03 mmol) in dioxane (12 mL) and water (12 mL) were added K$_2$CO$_3$ (2.06 mmol) and methylchloroformate (1.24 mmol) at room temperature. The solution was stirred for 2 h, dioxane was evaporated under vacuum, and the aqueous layer was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with brine, dried, and evaporated under reduced pressure. The residual oil was presorbed on silica gel and purified by column chromatography (15−30% EtOAc/hexanes) to yield corresponding carbamate.

Methyl-(2R,3R,4R,5R)-3-(benzo[d][1,3]dioxol-5-yl)-5-(tert-butyldiphenylsilyloxy)-1-hydroxy-4-(methoxymethoxy)hexan-2-ylcarbamate (13). Yield 65%, R_f 0.52 (60% EtOAc/hexanes); ^{1}H NMR (CDCl$_3$) δ 7.26−7.50 (m, 10H), 6.68 (d, *J* = 8.0 Hz, 1H), 6.49 (m, 2H), 5.90 (d, *J* = 19.0 Hz, 2H), 4.75 (m, 3H), 4.31 (m, 1H), 3.86 (m, 1H), 3.71 (d, *J* = 10.0 Hz, 1H), 3.64 (s, 3H), 3.43 (m, 1H), 3.39 (s, 3H), 3.35 (m, 1H), 1.00 (m, 3H), 0.96 (s, 9H); ^{13}C NMR (CDCl$_3$) δ 147.8, 146.7, 135.9, 135.8, 134.8, 134.1, 129.5, 127.5, 127.3, 122.9, 109.9, 108.4, 101.1, 98.9, 83.1, 70.1, 64.8, 56.4, 52.3, 46.1, 27.1, 19.9, 19.2; HRMS *m/z* (ESI) calcd for $C_{33}H_{43}NO_8SiNa$ (M + Na)$^+$ 632.2650, found 632.2656.

Methyl N-(1R+S,2R)-2-(1,3-Benzodioxol-5-yl)-1-(hydroxymethyl)-2-[(4R,5R)-2,2, 5-trimethyl-1,3-dioxolan-4-yl]ethylcarbamate (17a). Yield 86%; R_f 0.50 (60% EtOAc/hexanes); $[\alpha]^{22}_D$ −5.24 (*c* 0.56, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.54−6.82 (m, 3H), 5.94 (s, 2H), 4.12 (m, 1H), 3.78 (m, 2H), 3.64 (m, 3H), 3.54 (s, 3H), 2.82

(m, 1H), 1.42 (s, 6H), 0.76 (d, *J* = 5.8 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 156.7, 148.0, 147.1, 122.3, 122.2, 109.2, 108.6, 108.0, 101.3, 82.7, 64.1, 56.1, 52.3, 50.9, 27.4, 27.1, 18.9; HRMS *m/z* (ESI) calcd for $C_{18}H_{25}NO_7Na$ (M + Na)$^+$ 390.1528, found 390.1529.

Methyl N-(1R+S,2R)-1-(Hydroxymethyl)-2-(7-methoxy-1,3-benzodioxol-5-yl)-2-[(4R,5R)-2,2,5-trimethyl-1,3-dioxolan-4-yl]ethylcarbamate (17b). Yield 71%; R_f 0.47 (10% MeOH/CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.37 (m, 2H), 5.93 (s, 2H), 4.07−3.41 (m, 13H), 2.84 (m, 1H), 1.38 (m, 6H), 0.73 (d, *J* = 6.0 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 156.9, 149.3, 143.7, 134.8, 132.4, 108.4, 108.0, 103.1, 101.6, 83.3, 82.7, 64.0, 56.9, 52.3, 50.9, 27.4, 27.1, 18.7; HRMS *m/z* (ESI) calcd for $C_{19}H_{27}NO_8Na$ (M + Na)$^+$ 420.1634, found 420.1644.

General Procedure for Isopropylidene Deprotection. Dowex-50 acid resin (450 mg) was added to a solution of the corresponding carbamate (1.06 mmol) in MeOH (40 mL). The reaction mixture was refluxed for 10 h. The resin was removed from the mixture and washed with methanol (3 × 30 mL). The combined organic fractions were evaporated under reduced pressure to yield corresponding pure triol as a semisolid.

Methyl N-[(1R,2R,3R,4R)-2-(1,3-Benzodioxol-5-yl)-3,4-dihydroxy-1-(hydroxy-methyl)pentyl]carbamate (18a). Yield 56%; R_f 0.26 (60% EtOAc/hexanes); $[\alpha]^{22}_D$ +9.41 (*c* 0.26, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.59−6.74 (m, 3H), 5.92 (s, 2H), 5.16 (m, 1H), 4.28 (m, 1H), 3.69 (s, 3H), 3.42 (m, 3H), 3.07 (m, 1H). 1.11 (d, *J* = 5.5 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 158.8, 148.1, 146.9, 131.6, 122.3, 109.1, 108.8, 101.2, 73.9, 67.0, 64.6, 53.0, 50.0, 21.4; HRMS *m/z* (ESI) calcd for $C_{15}H_{21}NO_7Na$ (M + Na)$^+$ 350.1216, found 350.1217.

Methyl N-[(1R,2R,3R,4R)-3,4-Dihydroxy-1-(hydroxymethyl)-2-(7-methoxy-1,3-benzodioxol-5-yl)pentyl]carbamate (18b). Yield 50%; R_f 0.51 (20% MeOH/CHCl$_3$); $[\alpha]^{22}_D$ +9.6 (*c* 0.062, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.34 (br s, 2H), 5.94 (s, 2H), 5.09 (m, 1H), 4.29 (m, 1H), 3.88 (s, 3H), 3.72−3.30 (m, 6H), 3.68 (s, 3H), 3.08 (m, 1H). 1.13 (m, 3H); ^{13}C NMR (CDCl$_3$) δ 158.8, 149.3, 143.8, 134.5, 132.4, 109.2, 102.5, 101.7, 73.9, 67.0, 64.3, 57.0, 53.0, 50.3, 21.2; HRMS *m/z* (ESI) calcd for $C_{16}H_{23}NO_8Na$ (M + Na)$^+$ 380.1321, found 380.1326.

General Procedure for Acetyl Protection. To a mixture of the corresponding triol (0.61 mmol) and DMAP (3 mg, 0.024 mmol) in dry pyridine (5 mL) at 0 °C was added acetic anhydride (1.87 g, 18.44 mmol). The reaction mixture was stirred at room temperature for 12 h and diluted with water (50 mL). The aqueous layer was extracted with ether (5 × 20 mL). The combined organic layers were washed with water (4 × 15 mL) and brine, dried (MgSO$_4$), and evaporated under reduced pressure. The oily residue was presorbed on silica gel and purified by column chromatography (2−5% MeOH/CHCl$_3$) to yield corresponding triacetate.

(1R,2R,3R)-4-(Acetyloxy)-1-[(1R)-1-(acetyloxy)ethyl]-2-(1,3-benzodioxol-5-yl)-3-[(methoxycarbonyl)amino]butyl Acetate (19a). Yield 88%; R_f 0.47 (50% EtOAc/hexanes); $[\alpha]^{22}_D$ +58.33 (*c* 0.02, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.76 (d, *J* = 7.8 Hz, 1H), 6.58 (s, 1H), 6.52 (d, *J* = 7.8 Hz, 1H), 5.96 (s, 2H), 4.63 (m, 1H), 4.46 (m, 2H), 3.85 (dd, *J* = 11.0, 5.5 Hz, 1H), 3.65 (m, 3H), 3.19 (dd, *J* = 11.2, 0.4 Hz, 1H), 2.22 (s, 3H), 2.03 (s, 3H), 1.97 (s, 3H), 1.07 (d, *J* = 6.3 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 170.9, 169.8, 156.8, 148.6, 147.6, 128.6, 121.9, 109.2, 108.5, 101.5, 72.8, 69.4, 65.5, 52.6, 48.7, 46.6, 21.1, 20.8, 16.8; HRMS *m/z* (ESI) calcd for $C_{21}H_{27}NO_{10}Na$ (M + Na)$^+$ 476.1538, found 476.1533.

(1R,2R,3R)-4-(Acetyloxy)-1-[(1R)-1-(acetyloxy)ethyl]-2-(7-methoxy-1,3-benzodi-oxol-5-yl)-3-[(methoxycarbonyl)amino]butyl Acetate (19b). Yield 96%; R_f 0.73 (8% MeOH/CHCl$_3$); $[\alpha]^{22}_D$ +60.0 (*c* 0.02, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.26 (d, *J* = 1.6 Hz, 1H), 6.15 (d, *J* = 1.4 Hz, 1H), 5.93 (s, 2H), 5.32 (dd, *J* = 11.3, 1.6 Hz, 1H), 4.62 (qd, *J* = 6.3, 1.6 Hz, 1H), 4.52 (d, *J* = 11.0 Hz, 1H), 4.36 (m, Hz, 1H), 3.85 (m, 3H), 3.66−3.64 (m, 1H), 3.61 (s,

3H), 3.12 (dd, J = 11.3, 3.0 Hz, 1H), 2.19 (s, 3H), 2.00 (s, 3H), 1.95 (s, 3H), 1.05 (d, J = 6.6 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 170.8, 170.7, 169.7, 156.7, 149.8, 144.0, 135.2, 129.4, 109.1, 102.2, 101.7, 72.9, 69.5, 65.5, 57.2, 52.5, 46.9, 21.1, 20.9, 16.7; HRMS m/z (ESI) calcd for C$_{22}$H$_{29}$NO$_{11}$Na (M + Na)$^+$ 506.1638, found 506.1637.

Confirmation of Azide Stereochemistry. A mixture of azides **16a** was subjected to the reaction conditions described in the preparation of compounds **18a,b** to form lactones **20** and **21**.

(3R,4R,5R)-3-Azido-4-(benzo[d][1,3]dioxol-5-yl)-5-((S)-1-hydroxyethyl)dihydrofuran-2(3H)-one (20). R_f 0.59 (50% EtOAc/hexanes); [α]$^{22}_{\text{D}}$ +63.57 (c 0.03, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.68–6.73 (m, 3H), 6.02 (s, 2H), 4.41 (d, J = 11.6 Hz, 1H), 4.28 (dd, J = 9.91, 3.03 Hz, 1H), 3.74 (m, 1H), 3.47 (dd, J = 11.6 Hz, 1H), 1.24 (d, J = 6.6 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 171.5, 148.7, 147.9, 128.5, 121.7, 109.0, 107.6, 101.8, 85.6, 66.2, 64.8, 49.3, 19.9; HRMS m/z (ESI) calcd for C$_{13}$H$_{13}$N$_3$O$_5$Na 314.0752, found 314.0753.

(3S,4R,5R)-3-Azido-4-(benzo[d][1,3]dioxol-5-yl)-5-((S)-1-hydroxyethyl)dihydrofuran-2(3H)-one (21). R_f 0.54 (50% EtOAc/hexanes); [α]$^{22}_{\text{D}}$ −62.75 (c 0.005, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.68–6.73 (m, 3H), 5.97 (s, 2H), 4.63 (d, J = 8.3 Hz, 1H), 4.50 (dd, J = 5.23, 2.48 Hz, 1H), 3.90 (m, 1H), 3.73 (dd, J = 8.1, 5.2 Hz, 1H), 1.33 (d, J = 6.6 Hz, 3H); HRMS m/z (ESI) calcd for C$_{13}$H$_{13}$N$_3$O$_5$Na 314.0752, found 314.0755.

General Procedure for Imidate Formation. Method 1[16]. A 1.1 M solution of Tf$_2$O (0.677 mL, 0.74 mmol) in anhydrous CH$_2$Cl$_2$ was added over a period of 20 min to a solution of a corresponding carbamate (0.15 mmol) and DMAP (54.6 mg, 0.44 mmol) in CH$_2$Cl$_2$ (4.5 mL) at 0 °C. The reaction mixture was left to stir until complete consumption of the carbamate as monitored by TLC. On average complete consumption of the carbamate appeared to be between 2 and 6 h. During this period of time the reaction was kept in the ice-bath, but no further additions of ice were made. The reaction mixture was then diluted with EtOAc (20 mL) and poured into saturated aqueous NaHCO$_3$. The aqueous layer was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with 0.5 M HCl (1 × 2 mL) and brine, dried over Na$_2$SO$_4$, and concentrated. The crude product was presorbed on either silica gel or TLC plate and separated chromatographically to yield the corresponding imidate as a colorless oil.

Method 2[20]. Trifluoromethanesulfonic anhydride (64 μL, 0.39 mmol, 1.1 equiv) was added over period of 5 min to a solution of a corresponding carbamate (0.350 mmol, 1 equiv) and 2-chloropyridine (36 μL, 0.39 mmol, 1.1 equiv) in CH$_2$Cl$_2$ (1.8 mL) at −78 °C. The reaction mixture was left to stir for 5 min and then was placed in an ice–water bath for 15 min to warm to 0 °C and stirred at that temperature for 1 h. The mixture was quenched with 1 M NaOH (0.5 mL) and diluted with CH$_2$Cl$_2$ (5 mL). The aqueous layer was extracted with CH$_2$Cl$_2$ (3 × 10 mL). The combined organic layers were washed with brine (2 mL), dried over anhydrous Na$_2$SO$_4$, and concentrated. The crude product was separated chromatographically on alumina gel (5–30% EtOAc/hexanes) to yield a pale yellow fraction containing a corresponding imidate. The combined fractions were concentrated and additionally purified with preparative TLC (60–75% EtOAc/hexanes) to yield the corresponding imidates as a colorless oil.

(1R,2R)-2-(Acetyloxy)-2-(7R,8R)-7-[(acetyloxy)methyl]-5-Methoxy-7,8-dihydro[1,3]dioxolo[4,5-g]isoquinolin-8-yl-1-methylethyl Acetate (22). Yield 54% (method 1), 71% (method 2); R_f 0.31 (75% EtOAc/hexanes); [α]$^{22}_{\text{D}}$ −24.0 (c 0.02, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 7.13 (s, 1H), 6.49 (s, 1H), 5.98 (s, 2H), 4.95 (dd, J = 7.9, 3.0 Hz, 1H), 4.85 (dd, J = 6.3, 3.0 Hz, 1H), 4.13 (t, J = 5.5 Hz, 1H), 4.03 (td, J = 10.7, 5.2 Hz, 1H), 3.81 (s, 3H), 3.64 (dd, J = 10.7, 7.7 Hz, 1H), 3.00 (d, J = 7.9 Hz, 1H), 2.12 (s, 3H), 2.09 (s, 3H), 1.97 (s, 3H), 1.11 (d, J = 6.3 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ

170.8, 170.2, 169.9, 160.2, 150.1, 147.6, 130.2, 119.0, 109.3, 105.7, 101.7, 75.1, 69.6, 64.3, 53.8, 53.2, 39.3, 21.3, 20.9, 16.9; HRMS m/z (ESI) calcd for C$_{21}$H$_{26}$NO$_9$ (M + H)$^+$ 436.1608, found 436.1602.

Mixture of (1R,2R)-2-(Acetyloxy)-2-(6R,7R)-6-[(acetyloxy)methyl]-4,9-dimethoxy-6,7-dihydro[1,3]dioxolo[4,5-h]isoquinolin-7-yl-1-methylethyl Acetate (24) and (1R,2R)-2-(Acetyloxy)-2-(7R,8R)-7-[(acetyloxy)methyl]-4,5-dimethoxy-7,8-dihydro[1,3]dioxolo-[4,5-g]isoquinolin-8-yl-1-methylethyl Acetate (25). Yield 18%, 1:2 (method 1), 59%, 1:2 (method 2); R_f 0.24 (75% EtOAc/hexanes); ^{1}H NMR of minor regioisomer **24** (CDCl$_3$) δ 6.20 (s, 1H), 6.05 (d, J = 15.1 Hz, 2H), 4.92–4.78 (m, 2H), 4.13–3.95 (m, 2H), 3.89 (s, 3H), 3.85 (s, 3H), 3.73–3.59 (m, 1H), 2.95 (d, J = 9.1 Hz, 1H), 2.17 (s, 3H), 2.14 (s, 3H), 1.98 (s, 3H), 1.07 (m, 3H); ^{1}H NMR of major regioisomer **25** (CDCl$_3$) δ 6.25 (s, 1H), 5.97 (d, J = 3.8 Hz, 2H), 4.92 (dd, J = 9.1, 2.2 Hz, 1H), 4.81 (dd, J = 6.0, 2.5 Hz, 1H), 4.13–3.95 (m, 2H), 3.92 (s, 3H), 3.82 (s, 3H), 3.73–3.59 (m, 1H), 2.95 (d, J = 9.1 Hz, 1H), 2.15 (s, 3H), 2.13 (s, 3H), 1.98 (s, 3H), 1.09 (d, J = 6.0 Hz, 3H); ^{13}C NMR of major regioisomer **25** (CDCl$_3$) δ 170.8, 170.2, 169.9, 160.7, 151.0, 142.2, 138.9, 131.9, 108.5, 105.0, 102.5, 101.8, 74.0, 69.6, 63.8, 60.9, 53.3, 40.4, 21.3, 20.9, 16.8; HRMS m/z (ESI) calcd for C$_{22}$H$_{28}$NO$_{10}$ (M + H)$^+$ 466.1713, found 466.1722.

General Procedure for Conversion of Imidate to Amide. Method 1. A 1.3 M solution of trimethylsilylchloride (45.7 μL, 59.6 μmol) in dry acetonitrile was added to a stirred suspension of imidate (45.1 μmol) and anhydrous potassium iodide (45.1 μmol) in dry acetonitrile (1 mL). The resulting mixture was heated under reflux for 1 h. Water (2 mL) was added, and the acetonitrile was evaporated under reduced pressure. The aqueous layer was extracted with EtOAc (3 × 5 mL), and the combined organic layers were washed with an aqueous solution of sodium thiosulphate (5%, 1 × 2 mL) and brine, dried over Na$_2$SO$_4$, and concentrated. The residue was purified by flash column chromatography (60% EtOAc/hexanes) to afford pure product as an oil.

Method 2. The corresponding imidate (82 μmol) was dissolved in THF (2 mL) and 2 M HCl (0.2 mL) and stirred at room temperature for 5 h. The reaction mixture was neutralized with saturated aqueous NaHCO$_3$ and extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with brine, dried over Na$_2$SO$_4$, and concentrated in vacuo. The crude product was purified by flash column chromatography on silica gel to afford pure product as a white solid.

(1R,2R)-2-(Acetyloxy)-2-(7R,8R)-7-[(acetyloxy)methyl]-5-oxo-5,6,7,8-tetrahydro[1,3]dioxolo[4,5-g]isoquinolin-8-yl-1-methylethyl Acetate (23). Yield 84% (method 1), 78% (method 2); R_f 0.17 (75% EtOAc/hexanes); mp 194–196 °C (CH$_2$Cl$_2$/hexane); [α]$^{22}_{\text{D}}$ −40.0 (c 0.025, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 7.47 (s, 1H), 6.43 (s, 1H), 6.16 (s, 1H), 6.02 (s, 2H), 5.20 (dd, J = 8.8, 1.9 Hz, 1H), 4.70 (dd, J = 6.3, 2.5 Hz, 1H), 3.98 (dd, J = 10.7, 7.9 Hz, 1H), 3.89 (td, J = 11.3, 6.0 Hz, 1H), 3.67 (dd, J = 11.3, 5.8 Hz, 1H), 3.03 (d, J = 8.8 Hz, 1H), 2.15 (s, 3H), 2.14 (s, 3H), 2.02 (s, 3H), 1.00 (d, J = 6.3 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 170.6, 170.3, 170.0, 163.6, 151.5, 148.3, 129.9, 122.8, 108.6, 108.3, 102.1, 74.6, 69.4, 65.5, 50.0, 40.8, 21.4, 20.8, 16.8; HRMS m/z (ESI) calcd for C$_{20}$H$_{23}$NO$_9$Na (M + Na)$^+$ 444.1271, found 444.1278.

(1R,2R)-2-(Acetyloxy)-2-(7R,8R)-7-[(acetyloxy)methyl]-4-methoxy-5-oxo-5,6,7,8-tetrahydro[1,3]dioxolo[4,5-g]isoquinolin-8-yl-1-methylethyl Acetate (26). Yield 83% (method 2); R_f 0.10 (75% EtOAc/hexanes); [α]$^{22}_{\text{D}}$ −62.3 (c 0.061, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 6.15 (s, 1H), 6.13 (d, J = 9.9 Hz, 2H), 6.05 (d, J = 5.2 Hz, 1H), 5.20 (dd, J = 9.4, 2.2 Hz, 1H), 4.76 (qd, J = 6.9, 6.6, 1.6, 1.4 Hz, 1H), 3.99–3.85 (br m, 2H), 3.90 (s, 3H), 3.64 (m, 1H), 3.00 (d, J = 9.6 Hz, 1H), 2.17 (s, 6H), 2.03 (s, 3H), 1.09 (d, J = 6.3 Hz, 3H); ^{13}C NMR (CDCl$_3$) δ 170.7, 170.2, 170.1, 161.9, 149.7, 146.3, 136.4, 129.1, 108.3, 105.7, 103.7, 74.2, 69.3, 65.6, 56.6, 50.2, 41.2, 21.5, 20.8, 16.8; HRMS m/z (ESI) calcd for C$_{21}$H$_{25}$NO$_{10}$Na (M + Na)$^+$ 474.1376, found 474.1381.

(1*R*,2*R*)-2-(Acetyloxy)-2-(7*R*,8*R*)-7-[(acetyloxy)methyl]-4-Hydroxy-5-oxo-5,6,7,8-tetrahydro[1,3]dioxolo[4,5-*g*]isoquinolin-8-yl-1-methylethyl Acetate (27). Yield 95% (method 1); R_f 0.4 (75% EtOAc/hexanes); $[\alpha]^{22}_D$ −61.1 (*c* 0.085, CHCl₃); ¹H NMR (CDCl₃) δ 12.09 (br s, 1H), 6.28 (m, 1H), 6.07 (s, 1H), 6.03 (d, *J* = 6.6 Hz, 2H), 5.15 (dd, *J* = 8.8, 2.2 Hz, 1H), 4.66 (qd, *J* = 6.3, 6.0, 2.2, 1.9 Hz, 1H), 4.01 (m, 2H), 3.65 (m, 1H), 3.03 (d, *J* = 9.1 Hz, 1H), 2.16 (s, 3H), 2.14 (s, 3H), 2.04 (s, 3H), 1.10 (d, *J* = 6.3 Hz, 3H); ¹³C NMR (CDCl₃) δ 170.6, 170.4, 170.0, 168.2, 152.3, 146.2, 134.3, 130.0, 106.5, 102.6, 101.7, 74.5, 69.3, 65.2, 50.2, 40.8, 21.3, 20.8, 16.7; HRMS *m/z* (ESI) calcd for $C_{20}H_{23}NO_{10}Na$ (M + Na)⁺ 460.1220, found 460.1231.

General Procedure for Removing the Acetyl Protecting Group. Potassium carbonate (50 mg, 0.36 mmol) was added to a solution of a corresponding protected ring C analogue (0.045 mmol) in methanol (5 mL). The resulting suspension was stirred at room temperature until total consumption of the starting material (18 h). The reaction mixture was neutralized with acetic acid and concentrated to off-white residue, which was dissolved in 4:1 CH₂Cl₂/MeOH, adsorbed on TLC plate, and separated chromatographically (40% MeOH/CH₂Cl₂) to yield corresponding pure product as a white powder.

(7*R*,8*R*)-8-[(1*R*,2*R*)-1,2-Dihydroxypropyl]-7-(hydroxymethyl)-7,8-dihydro[1,3]dioxolo[4,5-*g*]isoquinolin-5(6*H*)-one (6). Yield 71%; R_f 0.3 (40% MeOH/CH₂Cl₂); $[\alpha]^{22}_D$ −111.1 (*c* 0.045, MeOH); ¹H NMR (DMSO-*d₆*) δ 8.26 (br s, 1H), 7.21 (s, 1H), 6.92 (s, 1H), 6.03 (s, 2H), 3.83 (t, *J* = 7.4, Hz, 1H), 3.36 (dd, *J* = 12.9, 5.5 Hz, 1H), 3.27 (td, *J* = 10.5, 6.3 Hz, 1H), 3.16−3.04 (m, 3H), 1.01 (d, *J* = 6.3 Hz, 3H); ¹³C NMR (DMSO-*d₆*) δ 163.4, 150.2, 146.9, 134.2, 123.6, 110.3, 106.7, 101.9, 74.6, 65.5, 63.3, 52.3, 41.8, 21.2; HRMS *m/z* (ESI) calcd for $C_{14}H_{17}NO_6Na$ (M + Na)⁺ 318.0954, found 318.0952.

(7*R*,8*R*)-8-[(1*R*,2*R*)-1,2-Dihydroxypropyl]-4-hydroxy-7-(hydroxymethyl)-7,8-dihydro[1,3]dioxolo[4,5-*g*]isoquinolin-5(6*H*)-one (5). Yield 83%; R_f 0.46 (20% MeOH/CH₂Cl₂); $[\alpha]^{22}_D$ −52.9 (*c* 0.068, MeOH); ¹H NMR (DMSO-*d₆*) δ 8.35 (m, 1H), 6.50 (s, 1H), 6.01 (d, *J* = 3.3 Hz, 2H), 3.88 (m, 1H), 3.36 (m, 1H), 3.31 (dd, *J* = 10.5, 6.6 Hz, 1H), 3.14 (td, *J* = 10.5, 8.3 Hz, 1H), 3.07 (s, 2H), 1.73−1.54 (br s, 4H), 1.02 (d, *J* = 6.0 Hz, 3H); ¹³C NMR (DMSO-*d₆*) δ 168.7, 151.7, 145.6, 134.9, 132.6, 107.1, 102.3, 102.1, 74.9, 65.5, 63.0, 52.3, 41.8, 21.2; HRMS *m/z* (ESI) calcd for $C_{14}H_{17}NO_7Na$ (M + Na)⁺ 334.0903, found 334.0911.

(1*R*,3a*R*,10b*R*)-1-Hydroxy-1,3a,4,10b-tetrahydro[1,3]dioxolo[4,5-*g*]furo[3,4-*c*]isoquinolin-5(3*H*)-one (7). To a solution of 6 (42 mg, 0.14 mmol) in methanol (2 mL) was added NaIO₄ (36.4 mg, 0.17 mmol) in one portion at 0 °C. The resulting suspension was vigorously stirred for 1 h at room temperature. The precipitate was filtered, and the solution was concentrated under reduced pressure. The residue was purified by preparative TLC (20% MeOH/CH₂Cl₂) to afford pure product 7 (28.6 mg, 82%) as a white powder; R_f 0.59 (20% MeOH/CH₂Cl₂); $[\alpha]^{22}_D$ −11.1 (*c* 0.018, MeOH); ¹H NMR (DMSO-*d₆*) δ 7.83 (s, 1H), 7.33 (s, 1H), 6.80 (s, 1H), 6.65 (br s, 1H), 6.08 (d, *J* = 8.81 Hz, 2H), 5.07 (t, *J* = 3.8, Hz, 1H), 4.26 (t, *J* = 3.8 Hz, 1H), 4.09 (dd, *J* = 9.1, 3.85 Hz, 1H), 3.88 (d, *J* = 8.81 Hz, 1H), 3.14 (t, *J* = 5.8 Hz, 1H); ¹³C NMR (DMSO-*d₆*) δ 163.2, 151.0, 147.5, 132.3, 121.5, 108.3, 107.1, 104.5, 102.2, 72.8, 55.0, 48.8; HRMS *m/z* (ESI) calcd for $C_{12}H_{11}NO_5Na$ (M + Na)⁺ 272.0535, found 272.0548.

Cell Culture. Human cervical cancer cell line HeLa (ATCC S3) cells were cultured in RPMI-1640 (Gibco) and supplemented with 10% FBS (Gibco), 100 mg/L penicillin G, 100 mg/L streptomycin, 1.0 mM sodium pyruvate, 1.5 g/L sodium bicarbonate, and 4.5 g/L glucose (all from Sigma) at 37 °C in a humidified atmosphere with 10% CO₂. MCF-7/AZ, a variant of the human mammary carcinoma cells MCF-7, were cultured in a mixture of DMEM and HAMF12 (50/50) (Invitrogen) supplemented with 250 IU/mL penicillin, 100 *μ*g/mL streptomycin, and 10% FBS.

MTT Assay. Cells were transferred to microtiter plates in 100 *μ*L of medium at a concentration of 2×10^4 cells/mL. HeLa and MCF-7/AZ cells were incubated for 24 h before treatment with the drugs to allow proper adhesion. Cells were treated with the panel of the test compounds and incubated for 48 h. Twenty microliters of MTT reagent (5 mg/mL) was added to each well and incubated further for 2 h. The resulting formazan crystals were dissolved in 100 *μ*L of DMSO, and the OD was determined at a wavelength of 490 nm. The experiments were repeated at least three times for each compound per cell line.

Supporting Information Available: General experimental section, X-ray data of 23, details of the computational work, and copies of ¹H and ¹³C NMR spectra for all new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.