New Bivalent PKC Ligands Linked by a Carbon Spacer: Enhancement in Binding Affinity

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Received April 29, 2003

Protein kinase C (PKC) is known to play an important role in many signal transduction pathways involved in hormone release, mitogenesis, and tumor promotion. In continuation of our efforts to find highly potent activators of PKC for possible use as Alzheimer's disease therapeutics, we designed and synthesized molecules containing two binding moieties (amides of benzolactams or esters of naphthylpyrrolidones) connected by a flexible spacer chain, which could theoretically bind to both the C1a and C1b activator binding domains of the catalytic region or to the C1 domains of two adjacent PKC molecules. The dimers **2a**− **g** of benzolactam showed a 200-fold increase in affinity to PKCα and -*δ* as the spacer length increased from 4 to 20 carbon atoms. Replacement of the oligomethylene chain with an oligoethylene glycol unit (compounds **2h**, **2i)** showed a 4000- to 7000-fold decrease in affinity to PKCα. The dimers of naphthylpyrrolidones **4a**−**g** did not show any marked improvement in binding affinities to PKC in comparison to the monomers synthesized earlier. The dimer of benzolactam **2e** did not show much selectivity for PKCα, -*β*I, -*δ*, -*ϵ*, and -*γ*. The high binding affinity of compounds **2d**−**g** to PKCs gives us the impetus to design additional molecules that would retain this enhanced activity and would also show selectivity for the PKC isoforms.

Introduction

Protein kinases C (PKCs) are serine/threonine kinases that transduce a myriad of signals to activate cellular functions. These include secretion, proliferation, differentiation, apoptosis, permeability, migration, and hypertrophy. At least 11 members of the PKC family have been identified. The isozymes are distributed in different tissues and play different physiological roles, and isozyme-specific modulators are of great current interest in the development of new medicinal leads and cancer therapeutics.[1−4] The structure of PKC is modular, consisting of a C-terminal catalytic region that functions as a serine/threonine kinase and an NH_2-terminal regulatory region that mediates membrane association and activation. PKC isozymes are classified according to the structural and functional differences in these regulatory domains. In the case of the "conventional" PKCα, -*β*I/*β*II, and -*γ* isozymes, the regulatory domain includes the activator-binding domains (C1) and the Ca^{2+}-binding domain C2. The C1 domains are present in a tandem C1a and C1b arrangement, each of which can potentially bind the endogenous activator diacylglycerol (DAG) and exogenous activators including phorbol esters. The "novel" PKC*δ*, -*ϵ*, -*η*, and -*θ* isozymes contain C2 domains that lack Ca^{2+} binding ability but retain functional C1a and C1b domains. The "atypical" PKCζ, -*ι*, and -*λ* regulatory domains also lack a functional C2 domain and contain a single C1 domain that does not bind diacylglycerol and whose regulation remains obscure. The protein kinases D (PKDs) *μ* and *ν* contain tandem C1a and C1b domains, which are, however, more widely separated than in the PKCs; the PKDs lack the C2 domain and have a pleckstrin homology domain. These and other differences cause the PKDs to be classified as a family distinct from the PKCs although related.

The C1 domains represent C3H1 zinc finger structures that bind diacylglycerol and its ultrapotent analogues such as the phorbol esters and indole alkaloids. X-ray crystallographic analysis of the complex consisting of the C1b domain of PKC*δ* and phorbol 13-acetate suggested a model that the C1 domain functions as a hydrophobic switch.[5] Ligand binds to a hydrophilic cleft in an otherwise hydrophobic surface. The enhanced hydrophobicity of this surface after binding, reflecting both the completion of the hydrophobic surface by the binding of the ligand itself together with further contributions from hydrophobic decorations on the ligand, promotes the interaction of the C1 domain with the phospholipid bilayer of the cell membranes and thereby drives removal of the pseudosubstrate region, adjacent to the C1 domain, from the catalytic site of the enzyme.

There is increasing evidence that the existence of two C1 domains affords complex modulatory opportunities in the regulation of PKC activity.[6−8] Expression of mutated PKCα in NIH3T3 cells and examination of the ability of ligands to induce translocation to the membrane showed that both domains play equivalent roles for translocation in response to phorbol 12-myristate 13-acetate (PMA), mezerein, and (−)-octylindolactam.[9] A similar approach showed different behavior for PKC*δ*.[10,11] For PKC*δ*, the ligands PMA, indolactam, and *n*-octyl-

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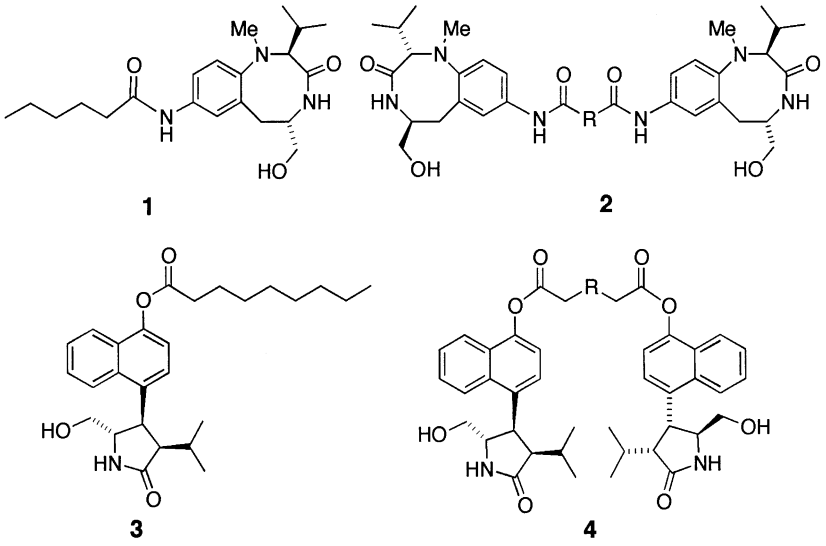
10.1021/jm0302041 CCC: $25.00
Published on Web 08/09/2003

Chart 1



indolactam were selectively dependent on the C1b domain, whereas selectivity was not seen for mezerein and bryostatin 1. Other approaches have also suggested that the C1a and C1b domains are not equivalent, although the apparent specific contributions of the domains differ depending on the specific methodological approach and on the response evaluated.[12−14] For example, mutants of PKC lacking the C1a or C1b domains, expressed in yeast, and evaluated for effects on cell growth suggest the nonequivalence of these domains, although the specific roles do not match those described above. Studies using binding of a fluorescent phorbol derivative suggested that there are two activator sites on PKC with low and high affinities and with distinct ligand selectivity. Analysis of phorbol ester binding using synthetic, refolded peptides corresponding to the C1a and C1b domains of the different PKC isoforms argued that the relative affinities of these two domains differed, depending on the isozyme, ranging from no activity for the C1a domain to activity equivalent to that of the C1b domain.[15,16] Studies with individual molecular constructs comprising single C1 domains likewise support lack of full equivalence.

Further differences are evident if the other receptors for DAG and phorbol esters with C1 domains are considered. In the PKDs,[17,18] the C1a domain is separated by a linker region of 80 residues compared to only 14−22 amino acids in the PKCs. Finally, the RasGRP family, the chimaerins, and the Munc proteins possess only a single C1 domain, in contrast to the PKCs and PKDs. Overall, these results strongly argue that the ligand-binding sites of the PKC isozymes do not follow a single pattern. There is therefore a strong incentive to look for highly potent and at the same time isozyme-selective PKC activators among molecules having two potential binding moieties linked by a flexible tether of variable length and appropriate nature. For such compounds, an enhanced binding affinity could be expected on the basis of the principle of additivity of the free energy of binding.[19]

In our previous work,[20] we have synthesized new benzolactam derivatives with an amide linkage containing saturated and unsaturated side chains and new naphthylpyrrolidone[21,22] derivatives with an ester linkage containing saturated and unsaturated side chains. We found that the nature of the side chain played a major role in the potency of these compounds. The stimulus to utilize these templates to explore the behavior of bidentate compounds stemmed from the ease of their synthesis and also from the fact that we had already established the activity of the monodentate compounds bearing a saturated ester- or amide-linked side chain.

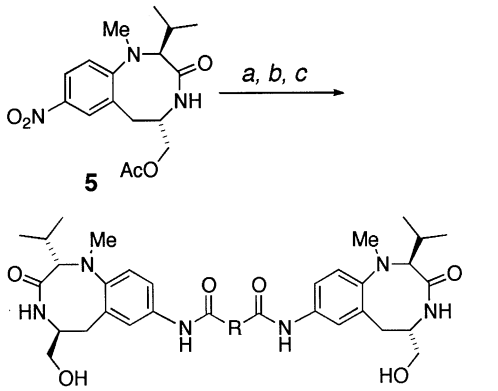
Design and Synthesis

In our earlier studies[20] we had shown that both saturated as well as unsaturated (dienic) side chains connected to benzolactams through an amide linkage gave rise to comparable binding affinities to PKCα. This prompted us to initially synthesize dimers of benzolactams with oligomethylene tethers of lengths differing from $n = 4$ to $n = 20$. Once the optimum chain length was established to lower the ClogP values of the derivatives, we also explored modification of the spacer by substituting the saturated chain with a poly(ethylene glycol) chain of similar length. To assess whether the realized binding affinity was due to the bidentate nature of the molecule, we also synthesized a monodentate molecule **7** with a benzolactam unit at one end of the chain and a 4-hydroxy naphthyl unit (which does not bind to PKC) at the other end.

The synthesis of the diamides **2a**−**i** (Chart 1, Scheme 1) started with the 8-nitrobenzolactam acetate (**5**), the synthesis of which has been previously published.[20,23] Catalytic reduction of the nitro group with Pd/C gave the intermediate 8-aminobenzolactam, which was utilized without purification for further reaction with the diacid chlorides. The diamides formed were then subjected to deacetylation with potassium carbonate in ethanol to form the required products in 18−38% yield over three steps. The saturated diacid chlorides were prepared by treatment of the respective diacids with an excess of thionyl chloride or oxalyl chloride.[24]

The monodentate derivative **7** of benzolactam was synthesized as shown in Scheme 2. The monomethyl ester of sebacic acid was refluxed with oxalyl chloride to obtain the acid chloride, which was reacted with

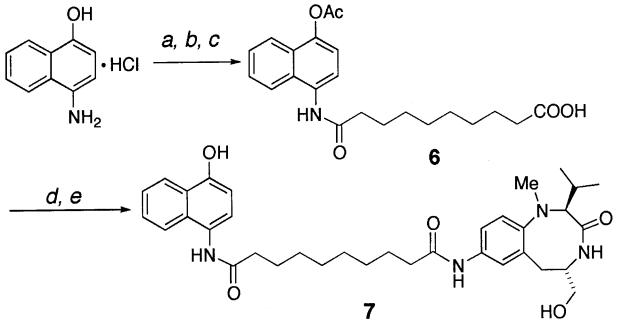
Scheme 1[a]



2a: R = -$(CH_2)_4$-
2b: R = -$(CH_2)_6$-
2c: R = -$(CH_2)_8$-
2d: R = -$(CH_2)_{10}$-
2e: R = -$(CH_2)_{12}$-
2f: R = -$(CH_2)_{14}$-
2g: R = -$(CH_2)_{20}$-
2h: R = -CH_2-$(OCH_2CH_2)_4O$-CH_2-
2i: R = -CH_2-$(OCH_2CH_2)_5O$-CH_2-

[a] Reagents and conditions: (a) Pd/C, H_2, MeOH, room temp; (b) Et_3N, diacid chloride, room temp; (c) Na_2CO_3, MeOH, room temp.

Scheme 2[a]



[a] Reagents and conditions: (a) methyl 9-(chlorocarbonyl-)nonanoate,[31] Et_3N, THF, room temp; (b) LiOH, MeOH, room temp; (c) Ac_2O, K_2CO_3, CH_2Cl_2; (d) 8-aminobenzolactam, Et_3N, room temp; (e) Na_2CO_3, MeOH, room temp.

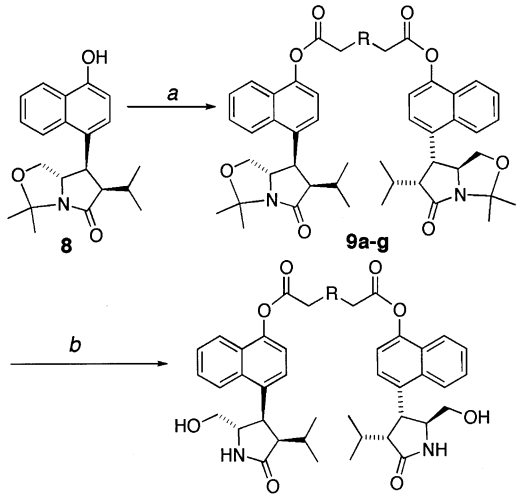
4-hydroxy-1-naphthylamine hydrochloride in the presence of triethylamine to give the amide. The methyl ester was then hydrolyzed with lithium hydroxide, and the resulting product was treated with acetic anhydride in the presence of 1 equiv of potassium carbonate to selectively protect the phenolic hydroxyl as the acetate. The free acid **6** was coupled with 8-aminobenzolactam in the presence of 1-[3′-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride to give the monodentate benzolactam derivative **7**.

The synthesis of the dimers of naphthylpyrrolidone **4a**−**g** was accomplished by the esterification of the parent phenols **8**[21] with the diacid chlorides. The esters **9** were subsequently subjected to acetonide cleavage by treatment with 1,2-ethanedithiol and boron trifluoride diethyl ether to give the required dimers (Scheme 3).

Binding Studies

The interaction of all dimers with PKC was appraised by evaluating their ability to displace [20-^{3}H]phorbol 12,13-dibutyrate (PDBU) binding from recombinant bovine PKCα, murine PKCδ, and the C1b domain of murine PKCδ. Preparation of these constructs has been described previously,[25,26] as has the [^{3}H]PDBu binding assay.[27] The results for the dimers of benzolactam are given in Table 1, and those for the dimers of naphthylpyrrolidones are given in Table 2. All compounds gave full inhibition of [^{3}H]PDBU binding. The K_i values of all analogues are in the nanomolar range, and some of the dimers of benzolactam show very high potency toward both PKCα and PKCδ. To test for isozyme

Scheme 3[a]



4a: R = -$(CH_2)_2$-
4b: R = -$(CH_2)_4$-
4c: R = -$(CH_2)_6$-
4d: R = -$(CH_2)_8$-
4e: R = -$(CH_2)_{10}$-
4f: R = -$(CH_2)_{12}$-
4g: R = -$(CH_2)_{18}$-

[a] Reagents and conditions: (a) diacid chloride, pyridine, room temp; (b) 1,2-ethanedithiol (10 equiv), BF_3•Et_2O (2 equiv), CH_2Cl_2, room temp.

Table 1. K_i and ClogP Values of Dimers **2a**−**i** and **4**[a]

compd	PKCα + Ca^{2+} K_i (nM)	PKCδ + Ca^{2+} K_i (nM)	C1b of PKCδ + Ca^{2+} K_i (nM)	ClogP[c]
1[b]	225 ± 16			6.0
2a	320 ± 60	67.8 ± 1.5	16.8 ± 0.64	2.77
2b	558 ± 27	170 ± 12	30.6 ± 4.8	4.65
2c	37.9 ± 1.2	5.75 ± 0.58	1.27 ± 0.30	5.71
2d	5.63 ± 0.56	1.31 ± 0.26	0.48 ± 0.04	6.76
2e	3.67 ± 0.23	0.78 ± 0.02	0.33 ± 0.06	7.82
2f	1.82 ± 0.34	0.36 ± 0.02	0.14 ± 0.013	8.88
2g	2.39 ± 0.30	0.38 ± 0.80	0.11 ± 0.008	12.05
2h	7860 ± 420			2.18
2i	4470 ± 320			2.04
7	56.6 ± 3.9	21.6 ± 1.5	4.4 ± 0.82	5.76

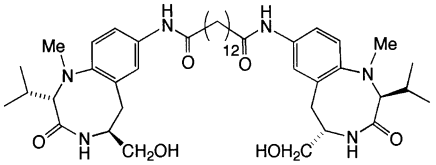
[a] K_i values are the mean ± SEM of three independent experiments. [b] Data taken from ref 20. [c] http://www.daylight.com/daycgi/clogp.

Table 2. K_i and ClogP Values of Dimers of Naphthylpyrrolidones **7a**−**g**[a]

compd	PKCα + Ca^{2+} K_i (nM)	PKCδ + Ca^{2+} K_i (nM)	ClogP[c]
3[b]	296 ± 12		5.76
4a	278 ± 8		4.47
4b	174 ± 7		5.53
4c	208 ± 33		6.59
4d	169 ± 3	44.8 ± 4.9	7.64
4e	154 ± 10		8.70
4f	254 ± 58		9.76
4g	296 ± 56		11.88

[a] K_i values are the mean ± SEM of three independent experiments. [b] Data taken from ref 22. [c] http://www.daylight.com/daycgi/clogp

Table 3. Human PKC Isozyme Data for Compound **2e**[a]



isozyme	K_d (nM)	K_i (nM)
PKCα + Ca^{2+}	0.28	0.93 ± 0.10
PKCβ-1 + Ca^{2+}	0.19	1.42 ± 0.20
PKCγ + Ca^{2+}	0.16	1.50 ± 0.30
PKCδ + EGTA	1.0	2.12 ± 0.21
PKCϵ + EGTA	0.37	0.55 ± 0.05

[a] K_i values are the mean ± SEM of three independent experiments.

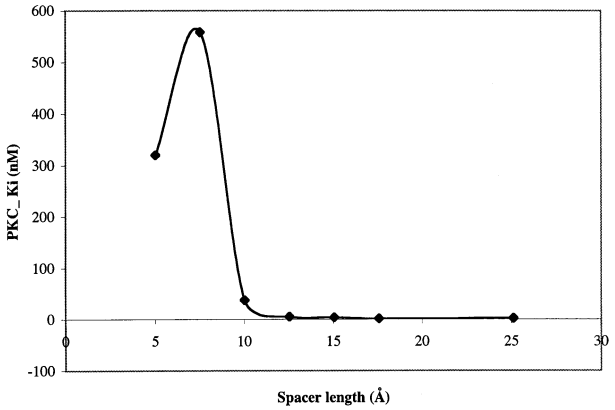


Figure 1. Correlation of binding affinities of **2a**−**g** with the spacer length in Å.

selectivity, the dimer **2e** was assessed for inhibitory activity against the human isoforms PKCα, PKCβI, PKCγ, PKCδ, and PKCϵ (Pan Vera, Madison, WI). The results are listed in Table 3. The K_i values for all PKCs were in the nanomolar range and comparable.

Discussion and Conclusions

In the case of the dimers of benzolactam, the nature of the spacer and its length play an important role in the potencies of the compounds toward the isozymes PKCα and PKCδ. This is clearly seen from the K_i values of **2a**−**i** (Table 1). A graphical representation between the correlation of spacer length and binding affinity to PKCα is depicted in Figure 1. The activities of **2a** and **2b** are slightly reduced compared to that of the reference compound **1**.[20] As the spacer length is increased from four to six carbons, there is an initial decrease in binding affinity. When the length is increased further by another two carbons, there is a dramatic enhancement in binding affinity (and a correspondingly smaller K_i). Marginal further enhancements are observed with additional increments in chain length from the 8-carbon chain to the 10-, 12-, and 14-carbon chain. For a 20-carbon chain, there is a slight decrease in affinity for PKCα, but the affinity for PKCδ remains unchanged. Indeed, the activities of **2d**, **2e**, **2f**, and **2g** are comparable, within 3-fold difference of each other. Compound **2f** has an almost 100-fold higher affinity for PKCα than does the *n*-pentylamide derivative **1**. All compounds showed a modest selectivity for PKCδ vs PKCα. In turn, the affinities for PKCδ paralleled the affinities for the C1b domain of PKCδ (C1b of PKCδ, Table 1). On the basis of the two-site model that depicts a single ligand molecule bound simultaneously to the two target sites C1a and C1b on a single molecule of PKC, the binding affinity of the dimer can be defined in terms of the binding affinities of individual ligands and a contribution by the spacer (S) to the binding affinity: K_i(dimer) = K_i(binding to C1a) K_i(binding to C1b) S.[19,28−30] The K_i value for binding to the C1b domain of the parent benzolactam (unsubstituted) is 334.0 ± 14.0 × 10^{-9} M.[5] Assuming a similar binding affinity to the C1a domain, the value for K_i(dimer) can be calculated as [334.0 × 10^{-9} M][334.0 × 10^{-9} M] = 1.0 × 10^{-13} M. For S = 1, the observed affinity is much weaker than the calculated one. From these results, we conclude that these dimers do not bind simultaneously to both C1 domains on the same molecule of protein kinase C, presumably because of steric constraints and that the compounds do not show a bidentate character. This is consistent with the result for the monodentate derivative **7**, which had affinities (within a 1- to 4-fold range) comparable to those of **2c** (having the same spacer length) for all three biological systems, namely, PKCα, PKCδ, and the C1 domain of PKCδ (Table 1). It should of course be noted that for a bidentate ligand each mole of ligand contains two moles of binding moiety, and potencies should therefore be corrected by a factor of 2 for appropriate comparison with a monodentate ligand. Since the bidentate compounds were able to fully compete for [^{3}H]-PDBu binding to PKCα and -δ, it is clear that they can bind to both the C1a and C1b domains of PKC and their failure to show the predicted enhancement in affinity cannot be explained by their activity on only one of these C1 domains.

The naphthylpyrrolidone dimers **4a**−**g** showed moderate activity in the high nanomolar range for PKCα (Table 2). The activity of these dimers was in the same range as that of our earlier synthesized monomer,[21] thus showing that the presence of two binding moieties in a single molecule did not yield any significant improvement. There is a slight improvement in activity as the length of the spacer is increased from 10- to 12- and 14-carbon atoms. A further increase in the chain length to 16- and 22-carbon atoms reduces the binding affinity. All differences in affinity are within a 2-fold range. The results from the dimers of both benzolactam and naphthylpyrrolidones illustrate that the presence of two binding moieties does not result in any synergistic binding with the C1 domains of PKCs.

Compound **2e** was selected for testing with different isozymes of PKC, namely, α, βI, δ, γ, and ϵ (Table 3). Its affinity was almost equal for all of these isozymes, with a slightly higher affinity for PKCϵ and lower affinity for PKCδ. Compounds **2f** and **2g** showed very high affinity toward the entire series and are comparable to PDBU (phorbol 12,13-dibutyrate). The derivatives **2h** and **2i**, in which the oligomethylene chain is replaced by an oligoethylene glycol chain, exhibit drastically reduced binding affinities (Table 1). This shows that the nature of the spacer is very important to achieve good binding to the PKCs.

It has been shown that the lipophilicity of molecules plays a crucial role in determining their biological activity and potency. In the case of phorbol esters, the phorbol 12,13-di-C8 esters and the 12-myristoyl deriva-

tive are among the most potent tumor promoters, whereas the 12-deoxy-13-phenylacetate derivative actually inhibits tumor promotion.[31−33] The length of the side chain is important for the ligand to rapidly equilibrate in aqueous solution and for the translocation of the complex to the plasma membrane.[34] In the case of phorbol esters, longer chain lengths resulted in loss of tumor-promoting activity due to nonequilibration in aqueous solution whereas very short chain lengths showed decrease in affinity for ligand added directly to the lipid phase. In the case of the dimers of the benzolactam, this relationship is not seen because **2e**, **2f**, and **2g** (containing 12C, 14C and 20C spacers) show very high binding affinity for both PKCα and PKCδ. It should be noted, however, that in these experiments the compounds were added directly to the lipids to avoid problems of equilibration. Little effect of linker length on activity was seen for the naphthylpyrrolidone dimers **4a**−**g**.

Even though compounds **2d**−**g** showed no isozyme specificity, their affinities were much higher than those reported earlier for the monomeric benzolactam amide series.[20] This increase in affinity could be due to the extensive hydrophobic facade presented by the spacer chain. It is generally accepted that hydrophobic moieties increase the ability of the compound to form a stable association with the membrane, which is known to be essential for inducing down-regulation of PKC. Earlier studies by our group[35] on both phorbol and lyngbyatoxin A have shown that the presence of a hydrophobic side chain (preferably 8−10 carbons) provides for a tight association of the enzyme to the membrane. In this connection, we note that certain lyngbyatoxin A analogues have been explored previously by us in which, for example, incorporation of a short, more polar 4-hydroxybutyl chain at the 7-position led to a reduction in PKC activation, presumably due to poorer membrane interaction. The ability of this compound to act as a PKC antagonist could not be observed under our experimental conditions.[36,37] More recently, Shibasaki and coworkers,[38,39] following a similar logic, have described a phorbol ester derivative comprising ethylene glycol units that is capable of acting as a weak PKC antagonist. This antagonist activity was again ascribed to reduced membrane interaction and inability to bring about PKC translocation. While the increased activity of the bivalent ligands **2d**−**g** described in this paper is likely due to more favorable membrane interaction as mentioned, we cannot rule out the possibility that the increase in affinity may also stem from additional interactions of the second binding unit with other sites on the protein. The present findings thus add to our storehouse of knowledge on ligand interactions with PKC. Such information will be valuable for achieving a better understanding of how these side chain appendages can be manipulated to influence certain cellular events coupled to PKC activation.

Experimental Section

Analysis of the Inhibition of [^{3}H]PDBU Binding by Nonradioactive Ligands. Enzyme−ligand interactions were analyzed by competition with [^{3}H]PDBU binding to the isozymes PKCα, -δ, -βI, -γ, and -ε as described previously.[25,40]

Chemistry. Thin-layer chromatography was performed in a solvent-vapor-saturated chamber on silica gel 60 F-254 plates. Spots were visualized by means of a UV lamp. Melting points (uncorrected) were determined in open capillaries on a Thomas-Hoover apparatus. ^{1}H and ^{13}C NMR spectra were recorded on a Varian instrument at 300 and 75 MHz, respectively, using TMS as an internal standard. Mass spectra were obtained on a Shimadzu QP-5000 mass spectrometer using a direct inlet probe and an electron beam energy of 70 eV. Determination of purity by HPLC was performed with a Shimadzu LC-10-AD system using the following conditions: (A) Discovery RP amide C_{16} 250 mm × 3.0 mm, flow rate = 0.5 mL/min, detection at 280 nm, 0−30 min, 40−80% acetonitrile in water, then 80% acetonitrile in water; (B) Discovery RP amide C_{16} 250 mm × 3.0 mm, flow rate = 0.5 mL/min, detection at 280 nm, 0−15 min, 0−100% methanol in water, then methanol; (C) Waters μ Bondapak C_{18} 300 mm × 7.8 mm, flow rate = 2.8 mL/min, detection at 280 nm, 0−30 min, 40−80% acetonitrile in water, then 80% acetonitrile in water.

General Procedure for the Synthesis of Compounds 2. The nitro compound **5** (100 mg, 0.287 mmol) was dissolved in ethanol (15 mL), and 10% Pd/C (15 mg, 0.014 mmol) was added to the reaction mixture. The mixture was vigorously stirred under a H_2 atmosphere overnight and then filtered, and the filtrate was concentrated to give a residue, which was dried under vacuum for 1 h at 60 °C. To a solution of the residue in anhydrous tetrahydrofuran (4 mL) under nitrogen was added anhydrous triethylamine (0.5 mL) and diacid chloride (0.143 mmol). The reaction mixture was stirred at room temperature for 4 h, then filtered and concentrated. Preparative TLC (10:1 ethyl acetate/ethanol as developing solvent) provided the acetyl-protected diamide, which was dissolved in ethanol (30 mL). A solution of sodium carbonate (60 mg) in water (6 mL) was added with vigorous stirring. After 1.5 h at room temperature, the solution was concentrated and an additional amount of water (20 mL) was added. Extraction with ethyl acetate (4 × 30 mL) provided the diamide with free hydroxyl groups, which was purified by preparative TLC (10:1 ethyl acetate/ethanol as developing solvent).

Hexanedioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*]-[1,4]diazocin-8-yl]amide (2a): yield 18%; $[\alpha]^{20}_D$ −249 (*c* 0.65, EtOH); IR (KBr) 3368, 1647, 1636, 1506 cm^{-1}; ^{1}H NMR (CD_3-OD) δ 0.98 (d, 6H, *J* = 6.8 Hz), 1.12 (d, 6H, *J* = 6.8 Hz), 1.71−1.78 (m, 4H), 2.31−2.44 (m, 6H), 2.72 (s, 3H), 2.81 (dd, 2H, *J* = 9.3, 15.9 Hz), 3.07 (dd, 2H, *J* = 4.4, 16.2 Hz), 3.38 (d, 2H, *J* = 6.6 Hz), 3.52 (dd, 2H, *J* = 6.8, 11.0 Hz), 3.59 (dd, 2H, *J* = 4.4, 11.2 Hz), 4.69−4.78 (m, 2H), 7.17 (d, 2H, *J* = 8.5 Hz), 7.30 (d, 2H, *J* = 2.4 Hz), 7.36 (dd, 2H, *J* = 2.4, 8.5 Hz); ^{13}C NMR (CD_3OD) δ 19.5, 21.3, 26.7, 30.1, 37.8, 38.5, 39.0, 54.7, 65.6, 77.4, 121.1, 123.9, 124.6, 135.7, 136.1, 150.0, 174.2, 175.9; MS *m*/*z* 319, 277, 149, 72, 45; HPLC retention time 6.3 min (98.1% purity) using conditions A, 29.2 min (98.5% purity) using conditions B.

Octanedioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*]-[1,4]diazocin-8-yl]amide (2b): yield 26%; $[\alpha]^{20}_D$ −208 (*c* 0.13, EtOH); IR (KBr) 3374, 1658, 1642, 1502 cm^{-1}; ^{1}H NMR (CD_3-OD) δ 0.98 (d, 6H, *J* = 6.8 Hz), 1.12 (d, 6H, *J* = 6.6 Hz), 1.37−1.45 (m, 4H), 1.69 (m, 4H), 2.33 (t, 4H, *J* = 7.2 Hz), 2.38 (m, 2H), 2.72 (s, 6H), 2.78 (dd, 2H, *J* = 9.5, 16.1 Hz), 3.07 (dd, 2H, *J* = 4.4, 16.1 Hz), 3.38 (d, 2H, *J* = 6.6 Hz), 3.51 (dd, 2H, *J* = 6.8, 11.0 Hz), 3.59 (dd, 2H, *J* = 4.9, 11.0 Hz), 4.74 (m, 2H), 7.16 (d, 2H, *J* = 8.5 Hz), 7.31 (d, 2H, *J* = 2.4 Hz), 7.36 (dd, 2H, *J* = 2.4, 8.5 Hz); ^{13}C NMR (CD_3OD) δ 19.5, 21.6, 26.2, 30.0, 30.1, 37.9, 38.5, 39.0, 54.7, 65.6, 77.4, 121.0, 123.8, 124.6, 135.7, 136.1, 149.9, 174.6, 175.8; MS *m*/*z* 558, 277, 159, 147, 72, 44; HPLC retention time 8.7 min (99.2% purity) using conditions A, 11.0 min (97.0% purity) using conditions C.

Decanedioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*]-[1,4]diazocin-8-yl]amide (2c): yield 38%; $[\alpha]^{20}_D$ −203 (*c* 0.55, MeOH); IR (KBr) 3387, 1657, 1642, 1503 cm^{-1}; ^{1}H NMR (CD_3-OD) δ 0.97 (d, 6H, *J* = 6.8 Hz), 1.11 (d, H, *J* = 6.6 Hz), 1.30−1.41 (m, 8H), 1.67 (m, 4H), 2.31 (t, 4H, *J* = 7.2 Hz), 2.38 (m, 2H), 2.71 (s, 6H), 2.78 (dd, 2H, *J* = 9.5, 16.1 Hz), 3.07 (dd, 2H,

J = 4.4, 16.1 Hz), 3.38 (d, 2H, J = 6.6 Hz), 3.51 (dd, 2H, J = 6.8, 11.0 Hz), 3.59 (dd, 2H, J = 4.6, 11.0 Hz), 4.74 (m, 2H), 7.16 (d, 2H, J = 8.5 Hz), 7.31 (d, 2H, J = 2.4 Hz), 7.36 (dd, 2H, J = 2.4, 8.5 Hz); ^{13}C NMR (CD_3OD) δ 19.5, 21.3, 27.0, 30.1, 30.3, 30.4, 38.0, 38.5, 39.0, 54.7, 65.6, 77.3, 121.0, 123.8, 124.5, 135.7, 136.1, 149.9, 174.6, 175.8; HPLC retention time 16.2 min (96.3% purity) using conditions A, 13.6 min (97.4% purity) using conditions C.

Dodecanedioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*][1,4]diazocin-8-yl]amide (2d): yield 23%; $[\alpha]^{20}_D$ −200 (*c* 0.54, EtOH); IR (KBr) 3368, 1653, 1541, 1507 cm^{-1}; ^{1}H NMR (CD_3OD) δ 0.97 (d, 6H, J = 6.8 Hz), 1.12 (d, 6H, J = 6.8 Hz), 1.26−1.42 (m, 12H), 1.66 (m, 4H), 2.32 (t, 4H, J = 7.3 Hz), 2.38 (m, 2H), 2.72 (s, 6H), 2.78 (dd, 2H, J = 9.5, 16.1 Hz), 3.07 (dd, 2H, J = 4.4, 16.1 Hz), 3.38 (d, 2H, J = 6.8 Hz), 3.52 (dd, 2H, J = 6.8, 11.0 Hz), 3.59 (dd, 2H, J = 4.4, 10.8 Hz), 4.74 (m, 2H), 7.16 (d, 2H, J = 8.5 Hz), 7.31 (d, 2H, J = 2.4 Hz), 7.36 (dd, 2H, J = 2.4, 8.5 Hz); ^{13}C NMR (CD_3OD) δ 19.5, 21.3, 27.1, 30.1, 30.4, 30.5, 30.6, 38.1, 38.5, 39.0, 54.7, 65.6, 77.4, 121.1, 123.8, 124.5, 135.7, 136.1, 149.9, 174.7, 175.9; MS *m*/*z* 664, 278, 160, 147, 72, 44; HPLC retention time 24.0 min (98.0% purity) using conditions A, 29.6 min (98.2% purity) using conditions B.

Tetradecanedioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*][1,4]diazocin-8-yl]amide (2e): yield 33%; $[\alpha]^{20}_D$ −162 (*c* 0.36, MeOH); IR (KBr) 3367, 1652, 1541, 1507 cm^{-1}; ^{1}H NMR (CD_3OD) δ 0.97 (d, 6H, J = 6.8 Hz), 1.11 (d, 6H, J = 6.6 Hz), 1.26−1.42 (m, 16H), 1.66 (m, 4H), 2.32 (t, 4H, J = 7.4 Hz), 2.39 (m, 2H), 2.72 (s, 6H), 2.78 (dd, 2H, J = 9.5, 16.1 Hz), 3.07 (dd, 2H, J = 4.2, 16.1 Hz), 3.38 (d, 2H, J = 6.6 Hz), 3.52 (dd, 2H, J = 6.8, 11.0 Hz), 3.59 (dd, 2H, J = 4.8, 11.1 Hz), 4.74 (m, 2H), 7.16 (d, 2H, J = 8.5 Hz), 7.31 (d, 2H, J = 2.4 Hz), 7.36 (dd, 2H, J = 2.2, 8.5 Hz); ^{13}C NMR (CD_3OD) δ 19.5, 21.3, 27.1, 30.1, 30.4, 30.6, 30.7, 30.8, 38.1, 38.5, 39.0, 54.7, 65.6, 77.4, 121.1, 123.8, 124.6, 135.7, 136.1, 149.9, 174.7, 175.8; HPLC retention time 23.6 min (98.7% purity) using conditions A, 30.7 min (98.5% purity) using conditions B.

Hexadecanedioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*][1,4]diazocin-8-yl]amide (2f): yield 26%; $[\alpha]^{20}_D$ −155.7 (*c* 0.415, MeOH); IR (KBr) 3368, 1650, 1542, 1506 cm^{-1}; ^{1}H NMR (CD_3OD) δ 0.97 (d, 6H, J = 6.6 Hz), 1.11 (d, 6H, J = 6.8 Hz), 1.25−1.40 (m, 20H), 1.67 (m, 4H), 2.32 (t, 4H, J = 7.4 Hz), 2.38 (m, 2H), 2.72 (s, 6H), 2.78 (dd, 2H, J = 9.8, 16.1 Hz), 3.07 (dd, 2H, J = 4.2, 16.1 Hz), 3.38 (d, 2H, J = 6.6 Hz), 3.52 (dd, 2H, J = 6.7, 11.1 Hz), 3.60 (dd, 2H, J = 4.8, 11.1 Hz), 4.74 (m, 2H), 7.16 (d, 2H, J = 8.5 Hz), 7.31 (d, 2H, J = 2.4 Hz), 7.36 (dd, 2H, J = 2.2, 8.5 Hz); ^{13}C NMR (CD_3OD) δ 19.5, 21.3, 27.1, 30.1, 30.4, 30.6, 30.7, 30.8, 30.9, 38.1, 38.5, 39.0, 54.7, 65.6, 77.4, 121.1, 123.8, 124.6, 135.7, 136.1, 149.9, 174.7, 175.8; HPLC retention time 33.1 min (96.6% purity) using conditions A, 32.2 min (99.9% purity) using conditions B.

Docosanedioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*][1,4]diazocin-8-yl]amide (2g): yield 28%; $[\alpha]^{20}_D$ −102.0 (*c* 0.15, MeOH); IR (KBr) 3368, 1643, 1546, 1504 cm^{-1}; ^{1}H NMR (CD_3OD) δ 0.97 (d, 6H, J = 6.8 Hz), 1.11 (d, 6H, J = 6.6 Hz), 1.24−1.40 (m, 32H), 1.67 (m, 4H), 2.32 (t, 4H, J = 7.4 Hz), 2.38 (m, 2H), 2.72 (s, 6H), 2.78 (dd, 2H, J = 10.1, 16.2 Hz), 3.07 (dd, 2H, J = 4.0, 16.0 Hz), 3.38 (d, 2H, J = 6.6 Hz), 3.52 (dd, 2H, J = 6.8, 11.0 Hz), 3.60 (dd, 2H, J = 4.8, 10.9 Hz), 4.73 (m, 2H), 7.16 (d, 2H, J = 8.5 Hz), 7.31 (d, 2H, J = 2.4 Hz), 7.36 (dd, 2H, J = 2.2, 8.5 Hz); ^{13}C NMR (CD_3OD) δ 19.5, 21.3, 27.1, 30.1, 30.5, 30.6, 30.8, 30.9 (five overlapping signals), 38.1, 38.5, 39.0, 54.7, 65.6, 77.3, 121.1, 123.8, 124.6, 135.7, 136.1, 149.9, 174.7, 175.8; HPLC retention time 50.0 min (96.0% purity) using conditions A, 34.4 min (97.9% purity) using conditions B.

3,6,9,12,15-Pentaoxaheptadecane-1,17-dioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*][1,4]diazocin-8-yl]amide (2h): yield 21%; $[\alpha]^{20}_D$ −93 (*c* 0.55, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 0.99 (d, 6H, J = 6.8 Hz), 1.11 (d, 6H, J = 6.8 Hz), 2.31−2.4 (m, 2H), 2.66 (s, 6H), 2.84 (dd, 2H, J = 9.5, 15.6 Hz), 2.97 (dd, 2H, J = 5.4, 15.4 Hz), 3.25−3.31 (m, 4H), 3.4−3.46 (m, 4H), 3.57−3.74 (m, 14H), 3.78−3.88 (m, 2H), 4.02 (s, 2H), 4.03 (s, 2H), 4.73 (br s, 2H), 7.03 (d, 2H, J = 2.4 Hz), 7.14 (d, 2H, J = 8.5 Hz), 7.43 (d, 2H, J = 6.6 Hz), 7.6 (dd, 2H, J = 2.4, 8.5 Hz), 9.06 (br s, 2H). ^{13}C NMR ($CDCl_3$) δ 8.6, 20.8, 28.8, 29.7, 36.7, 38.5, 52.8, 64.4, 70.2, 70.3, 70.6, 71.1, 71.5, 120.2, 123.2, 123.7, 133.8, 135.1, 148.9, 168.4, 174.1. HPLC retention time 9.8 min (99.9% purity) using conditions A, 10.3 min (97.8% purity) using conditions C; HRMS calcd for ($C_{42}H_{64}N_6O_{11}$ + H^+) 829.4711, found 829.4695.

3,6,9,12,15,18-Hexaoxadocosane-1,20-dioic acid bis[*N*-(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*][1,4]diazocin-8-yl]amide (2i): yield 30%; $[\alpha]^{20}_D$ −154 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 1.00 (d, 6H, J = 6.8 Hz), 1.09 (d, 6H, J = 6.8 Hz), 2.27−2.41 (m, 2H), 2.65 (s, 6H), 2.69 (dd, 2H, J = 8.8, 18.1 Hz), 2.85 (dd, 2H, J = 9.7, 15.8 Hz), 2.95−3.08 (m, 4H), 3.21−3.4 (m, 4H), 3.38−3.61 (m, 20H), 4.00 (d, 2H, J = 16.1 Hz), 4.13 (d, 2H, J = 16.3 Hz), 4.83 (br s, 2H), 7.04 (d, 2H, J = 2.2 Hz), 7.14 (d, 2H, J = 8.6 Hz), 7.56 (d, 2H, J = 7.6 Hz), 7.64 (dd, 2H, J = 2.4, 8.6 Hz), 9.06 (s, 2H); ^{13}C NMR ($CDCl_3$) δ 18.5, 20.9, 28.9, 36.7, 38.8, 52.9, 64.4, 69.7, 69.9, 70.2, 70.3, 70.8, 71.2, 120.7, 123.3, 124.2, 134.1, 135.1, 148.9, 168.6, 174.0; HPLC retention time 15.5 min (96.5% purity) using conditions A, 17.0 min (98.6% purity) using conditions C; HRMS calcd for ($C_{44}H_{68}N_6O_{12}$ + H^+) 873.4973, found 873.4942.

General Procedure for the Preparation of the Dimers 9. To a solution of compound **8** (45.0 mg, 0.133 mmol) in pyridine (0.5 mL) and CH_2Cl_2 (0.5 mL) was added diacyl chloride (0.060 mmol). The reaction mixture was stirred under N_2 at room temperature for 36 h. Then the resulting solution was diluted with ethyl acetate (80 mL), washed with 1 N HCl (5 mL × 2), saturated aqueous $NaHCO_3$, and brine, dried over anhydrous Na_2SO_4, and concentrated. The residue was chromatographed on silica gel (25:1 CH_2Cl_2/acetone) to afford the dimer and recovered compound **8**.

Hexanedioic acid bis[4-[(6*R*,7*R*,7a*S*)-5,6,7,7a-tetrahydro-(6-isopropyl-3,3-dimethyl-5-oxo-1*H*-pyrrolo[1,2-*c*]oxazol-7-yl)]-1-naphthyl] ester (9a): yield 69%; $[\alpha]_D$ +287 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 0.80 (d, 6H, J = 7.2 Hz), 0.87 (d, 6H, J = 6.6 Hz), 1.34−1.45 (m, 2H), 1.60 (s, 6H), 1.79 (s, 6H), 2.03−2.12 (m, 4H), 2.84−2.94 (m, 4H), 3.25 (dd, 2H, J = 8.7, 3.6 Hz), 3.54 (t, 2H, J = 9.0 Hz), 4.15 (t, 2H, J = 9.0 Hz), 4.22 (dd, 2H, J = 8.7, 5.7 Hz), 4.99 (td, 2H, J = 9.3, 5.7 Hz), 7.26 (d, 2H, J = 8.1 Hz), 7.49 (d, 2H, J = 8.1 Hz), 7.53−7.63 (m, 4H), 7.93−7.99 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 18.9, 22.5, 24.0, 24.7, 27.1, 27.3, 34.2, 46.3, 59.6, 62.7, 69.9, 91.8, 117.5, 122.3, 123.4, 123.6, 126.9, 127.3, 127.5, 130.9, 133.4, 146.4, 171.4, 171.9.

Octanedioic acid bis[4-[(6*R*,7*R*,7a*S*)-5,6,7,7a-tetrahydro-(6-isopropyl-3,3-dimethyl-5-oxo-1*H*-pyrrolo[1,2-*c*]oxazol-7-yl)]-1-naphthyl] ester (9b): yield 48%; $[\alpha]_D$ +251 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 0.79 (d, 6H, J = 6.9 Hz), 0.87 (d, 6H, J = 6.9 Hz), 1.33−1.45 (m, 2H), 1.55−1.67 (m, 10H), 1.78 (s, 6H), 1.87−2.00 (m, 4H), 2.80 (t, 4H, J = 7.5 Hz), 3.25 (dd, 2H, J = 8.7, 3.3 Hz), 3.54 (t, 2H, J = 8.7 Hz), 4.14 (t, 2H, J = 9.3 Hz), 4.21 (dd, 2H, J = 8.4, 5.7 Hz), 4.99 (td, 2H, J = 9.3, 5.7 Hz), 7.25 (d, 2H, J = 8.4 Hz), 7.48 (d, 2H, J = 7.8 Hz), 7.53−7.63 (m, 4H), 7.92−7.99 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 18.9, 22.5, 24.0, 25.0, 27.0, 27.3, 29.1, 34.5, 46.3, 59.6, 62.7, 69.9, 91.8, 117.5, 122.3, 123.4, 123.6, 126.8, 127.4, 127.5, 130.7, 133.4, 146.5, 171.4, 172.30.

Decanedioic acid bis[4-[(6*R*,7*R*,7a*S*)-5,6,7,7a-tetrahydro-(6-isopropyl-3,3-dimethyl-5-oxo-1*H*-pyrrolo[1,2-*c*]oxazol-7-yl)]-1-naphthyl] ester (9c): yield 61%; $[\alpha]_D$ +217 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 0.79 (d, 6H, J = 6.9 Hz), 0.87 (d, 6H, J = 6.6 Hz), 1.32−1.59 (m, 10H), 1.60 (s, 6H), 1.79 (s, 6H), 1.82−1.96 (m, 4H), 2.76 (t, 4H, J = 7.5 Hz), 3.25 (dd, 2H, J = 8.7, 3.6 Hz), 3.54 (t, 2H, J = 9.0 Hz), 4.15 (t, 2H, J = 9.0 Hz), 4.22 (dd, 2H, J = 8.7, 5.7 Hz), 4.99 (td, 2H, J = 9.0, 5.7 Hz), 7.24 (d, 2H, J = 7.8 Hz), 7.48 (d, 2H, J = 8.1 Hz), 7.53−7.63 (m, 4H), 7.92−7.99 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 18.9, 22.5,

24.0, 25.2, 27.0, 27.3, 29.4, 34.6, 46.3, 59.6, 62.7, 69.9, 91.8, 117.5, 122.3, 123.4, 123.6, 126.8, 127.4, 127.5, 130.7, 133.4, 146.5, 171.5, 172.4.

Dodecanedioic acid bis[4-[(6*R*,7*R*,7a*S*)-5,6,7,7a-tetrahydro-(6-isopropyl-3,3-dimethyl-5-oxo-1*H*-pyrrolo[1,2-*c*]oxazol-7-yl)]-1-naphthyl] ester (9d): yield 53%; $[\alpha]_D$ +292 (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.79 (d, 6H, *J* = 6.9 Hz), 0.87 (d, 6H, *J* = 6.6 Hz), 1.33−1.56 (m, 14H), 1.60 (s, 6H), 1.79 (s, 6H), 1.80−1.93 (m, 4H), 2.75 (t, 4H, *J* = 7.5 Hz), 3.24 (dd, 2H, *J* = 8.7, 3.6 Hz), 3.53 (t, 2H, *J* = 8.7 Hz), 4.14 (t, 2H, *J* = 9.3 Hz), 4.21 (dd, 2H, *J* = 8.7, 5.7 Hz), 4.99 (td, 2H, *J* = 9.3, 5.7 Hz), 7.24 (d, 2H, *J* = 7.8 Hz), 7.48 (d, 2H, *J* = 7.8 Hz), 7.53−7.63 (m, 4H), 7.92−7.99 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 18.8, 22.5, 24.0, 25.3, 27.0, 27.3, 29.4, 29.5, 29.6, 34.6, 46.3, 59.6, 62.7, 69.9, 91.8, 117.5, 122.4, 123.4, 123.6, 126.8, 127.5, 130.7, 133.4, 146.5, 171.4, 172.5.

Tetradecanedioic acid bis[4-[(6*R*,7*R*,7a*S*)-5,6,7,7a-tetrahydro-(6-isopropyl-3,3-dimethyl-5-oxo-1*H*-pyrrolo[1,2-*c*]oxazol-7-yl)]-1-naphthyl] ester (9e): yield 41%; $[\alpha]_D$ +241 (*c* 0.7, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.79 (d, 6H, *J* = 7.2 Hz), 0.87 (d, 6H, *J* = 6.9 Hz), 1.29−1.54 (m, 18H), 1.60 (s, 6H), 1.78 (s, 6H), 1.80−1.92 (m, 4H), 2.75 (t, 4H, *J* = 7.5 Hz), 3.24 (dd, 2H, *J* = 8.7, 3.6 Hz), 3.53 (t, 2H, *J* = 8.7 Hz), 4.14 (t, 2H, *J* = 9.3 Hz), 4.21 (dd, 2H, *J* = 8.7, 5.7 Hz), 4.99 (td, 2H, *J* = 9.3, 5.7 Hz), 7.24 (d, 2H, *J* = 7.8 Hz), 7.48 (d, 2H, *J* = 7.8 Hz), 7.53−7.63 (m, 4H), 7.92−7.99 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 18.9, 22.5, 24.0, 25.3, 27.1, 27.3, 29.4, 29.5, 29.7, 29.8, 34.6, 46.3, 59.6, 62.7, 69.9, 91.8, 117.5, 122.4, 123.4, 123.6, 126.8, 127.5, 130.7, 133.4, 146.5, 171.5, 172.50.

Hexadecanenedioic acid bis[4-[(6*R*,7*R*,7a*S*)-5,6,7,7a-tetrahydro-(6-isopropyl-3,3-dimethyl-5-oxo-1*H*-pyrrolo-[1,2-*c*]oxazol-7-yl)]-1-naphthyl] ester (9f): yield 56%; $[\alpha]_D$ +213 (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.79 (d, 6H, *J* = 6.9 Hz), 0.87 (d, 6H, *J* = 6.6 Hz), 1.25−1.55 (m, 22H), 1.60 (s, 6H), 1.79 (s, 6H), 1.80−1.92 (m, 4H), 2.75 (t, 4H, *J* = 7.5 Hz), 3.24 (dd, 2H, *J* = 8.7, 3.6 Hz), 3.54 (t, 2H, *J* = 8.7 Hz), 4.14 (t, 2H, *J* = 9.0 Hz), 4.21 (dd, 2H, *J* = 8.7, 5.7 Hz), 4.99 (td, 2H, *J* = 9.0, 5.7 Hz), 7.24 (d, 2H, *J* = 7.8 Hz), 7.48 (d, 2H, *J* = 7.8 Hz), 7.53−7.63 (m, 4H), 7.92−7.99 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 18.8, 22.5, 24.0, 25.3, 27.0, 27.3, 29.4, 29.5, 29.7, 29.8, 29.9, 34.6, 46.3, 59.6, 62.7, 69.9, 91.7, 117.5, 122.4, 123.4, 123.6, 126.8, 127.4, 130.7, 133.4, 146.5, 171.4, 172.5.

Docosanedioic acid bis[4-[(6*R*,7*R*,7a*S*)-5,6,7,7a-tetrahydro-(6-isopropyl-3,3-dimethyl-5-oxo-1*H*-pyrrolo[1,2-*c*]oxazol-7-yl)]-1-naphthyl] ester (9g): yield 49%; $[\alpha]_D$ +209 (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 0.79 (d, 6H, *J* = 6.9 Hz), 0.87 (d, 6H, *J* = 6.6 Hz), 1.20−1.55 (m, 34H), 1.60 (s, 6H), 1.79 (s, 6H), 1.80−1.92 (m, 4H), 2.75 (t, 4H, *J* = 7.5 Hz), 3.25 (dd, 2H, *J* = 8.7, 3.6 Hz), 3.54 (t, 2H, *J* = 8.7 Hz), 4.15 (t, 2H, *J* = 9.0 Hz), 4.21 (dd, 2H, *J* = 8.7, 5.7 Hz), 4.99 (td, 2H, *J* = 9.3, 5.4 Hz), 7.24 (d, 2H, *J* = 7.8 Hz), 7.48 (d, 2H, *J* = 8.1 Hz), 7.53−7.63 (m, 4H), 7.92−7.99 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 18.9, 22.5, 24.0, 25.3, 27.0, 27.3, 29.4, 29.5, 29.7, 29.8, 29.9, 30.0, 34.6, 46.3, 59.6, 62.7, 69.9, 91.8, 117.5, 122.4, 123.4, 123.6, 126.8, 127.4, 130.6, 133.4, 146.5, 171.5, 172.5.

General Procedure for the Preparation of 4. To the solution of 25 μmol of **9** in anhydrous methylene chloride (3 mL) was added 1,2-ethanedithiol (65 μL) and $BF_3{\cdot}Et_2O$ (27 μL) at room temperature under N_2. After 10 min, the reaction was quenched by adding saturated aqueous $NaHCO_3$ (2 mL) and diluted with ethyl acetate (80 mL). The organic layer was washed with brine and dried over anhydrous Na_2SO_4. After concentration, the residue was chromatographed on silica gel (10:1 EtOAc/MeOH) to afford the product.

Hexanedioic acid bis[4-[(2*S*,3*R*,4*R*)-2-(hydroxymethyl)-4-isopropyl-5-oxo-3-pyrrolidinyl]-1-naphthalenyl] ester (4a): yield 93%; $[\alpha]_D$ +188 (*c* 1.0, MeOH); ^1H NMR (CD_3OD) δ 0.78 (d, 6H, *J* = 6.9 Hz), 0.84 (d, 6H, *J* = 6.6 Hz), 1.25−1.40 (m, 2H), 2.00−2.08 (m, 4H), 2.88−3.00 (m, 6H), 3.53 (dd, 2H, *J* = 11.4, 5.1 Hz), 3.78 (dd, 2H, *J* = 11.4, 3.0 Hz), 4.31−4.42 (m, 4H), 7.26 (d, 2H, *J* = 7.8 Hz), 7.50−7.65 (m, 6H), 7.97 (dd, 2H, *J* = 8.4, 1.2 Hz), 8.18 (d, 2H, *J* = 8.4 Hz); ^{13}C NMR (CD_3OD) δ 19.3, 23.0, 25.7, 28.3, 34.8, 43.1, 53.1, 60.3, 64.3, 118.7, 123.2, 125.0, 125.1, 127.7, 128.3, 128.8, 133.1, 135.1, 147.7, 173.9, 180.3. Anal. ($C_{42}H_{48}N_2O_8{\cdot}1.25H_2O$) C, H, N.

Octanedioic acid bis[4-[(2*S*,3*R*,4*R*)-2-(hydroxymethyl)-4-isopropyl-5-oxo-3-pyrrolidinyl]-1-naphthalenyl] ester (4b): yield 95%; $[\alpha]_D$ +192 (*c* 0.9, MeOH); ^1H NMR (CD_3OD) δ 0.77 (d, 6H, *J* = 6.9 Hz), 0.83 (d, 6H, *J* = 6.9 Hz), 1.28−1.38 (m, 2H), 1.57−1.67 (m, 4H), 1.84−1.97 (m, 4H), 2.83 (t, 4H, *J* = 7.5 Hz), 2.96 (dd, 2H, *J* = 9.0, 3.9 Hz), 3.52 (dd, 2H, *J* = 11.7, 5.1 Hz), 3.77 (dd, 2H, *J* = 11.7, 2.7 Hz), 4.30−4.42 (m, 4H), 7.24 (d, 2H, *J* = 7.8 Hz), 7.52−7.64 (m, 6H), 7.94 (dd, 2H, *J* = 8.1, 1.5 Hz), 8.17 (d, 2H, *J* = 7.8 Hz); ^{13}C NMR (CD_3OD) δ 19.3, 23.0, 26.0, 28.3, 30.0, 35.0, 43.1, 53.1, 60.3, 64.3, 118.7, 123.2, 125.0, 125.1, 127.7, 128.3, 128.8, 133.1, 135.1, 147.6, 174.2, 180.3. Anal. ($C_{44}H_{52}N_2O_8{\cdot}1.5H_2O$) C, H, N.

Decanedioic acid bis[4-[(2*S*,3*R*,4*R*)-2-(hydroxymethyl)-4-isopropyl-5-oxo-3-pyrrolidinyl]-1-naphthalenyl] ester (4c): yield 91%; $[\alpha]_D$ +186 (*c* 0.7, MeOH); ^1H NMR (CD_3OD) δ 0.77 (d, 6H, *J* = 6.9 Hz), 0.83 (d, 6H, *J* = 6.9 Hz), 1.27−1.39 (m, 2H), 1.44−1.60 (m, 8H), 1.80−1.92 (m, 4H), 2.79 (t, 4H, *J* = 7.5 Hz), 2.96 (dd, 2H, *J* = 9.0, 3.9 Hz), 3.52 (dd, 2H, *J* = 11.7, 5.4 Hz), 3.77 (dd, 2H, *J* = 11.7, 2.7 Hz), 4.30−4.42 (m, 4H), 7.23 (d, 2H, *J* = 7.8 Hz), 7.53−7.64 (m, 6H), 7.93 (dd, 2H, *J* = 7.8, 1.8 Hz), 8.17 (d, 2H, *J* = 7.8 Hz); ^{13}C NMR (CD_3OD) δ 19.3, 23.0, 26.2, 28.3, 30.3, 30.4, 35.1, 43.1, 53.1, 60.3, 64.3, 118.7, 123.2, 125.0, 125.1, 127.7, 128.3, 128.8, 133.1, 135.1, 147.7, 174.2, 180.3. Anal. ($C_{46}H_{56}N_2O_8{\cdot}0.5H_2O$) C, H, N.

Dodecanedioic acid bis[4-[(2*S*,3*R*,4*R*)-2-(hydroxymethyl)-4-isopropyl-5-oxo-3-pyrrolidinyl]-1-naphthalenyl] ester (4d): yield 89%; $[\alpha]_D$ +194 (*c* 0.7, MeOH); ^1H NMR (CD_3OD) δ 0.77 (d, 6H, *J* = 6.6 Hz), 0.83 (d, 6H, *J* = 6.9 Hz), 1.26−1.58 (m, 14H), 1.78−1.88 (m, 4H), 2.77 (t, 4H, *J* = 7.5 Hz), 2.96 (dd, 2H, *J* = 9.0, 3.9 Hz), 3.52 (dd, 2H, *J* = 11.7, 5.4 Hz), 3.77 (dd, 2H, *J* = 11.7, 2.7 Hz), 4.30−4.42 (m, 4H), 7.23 (d, 2H, *J* = 7.8 Hz), 7.53−7.64 (m, 6H), 7.93 (dd, 2H, *J* = 7.8, 1.5 Hz), 8.17 (d, 2H, *J* = 7.8 Hz); ^{13}C NMR (CD_3OD) δ 19.3, 23.0, 26.2, 28.3, 30.4, 30.5, 30.7, 35.1, 43.1, 53.1, 60.3, 64.3, 118.7, 123.2, 125.0, 125.1, 127.7, 128.3, 128.8, 133.1, 135.1, 147.7, 174.2, 180.3. Anal. ($C_{48}H_{60}N_2O_8{\cdot}0.75H_2O$) C, H, N.

Tetradecanedioic acid bis[4-[(2*S*,3*R*,4*R*)-2-(hydroxymethyl)-4-isopropyl-5-oxo-3-pyrrolidinyl]-1-naphthalenyl] ester (4e): yield 93%; $[\alpha]_D$ +191 (*c* 0.4, MeOH); ^1H NMR (CD_3OD) δ 0.77 (d, 6H, *J* = 6.6 Hz), 0.83 (d, 6H, *J* = 6.9 Hz), 1.26−1.56 (m, 18H), 1.77−1.88 (m, 4H), 2.77 (t, 4H, *J* = 7.5 Hz), 2.96 (dd, 2H, *J* = 9.6, 3.9 Hz), 3.52 (dd, 2H, *J* = 11.7, 5.1 Hz), 3.77 (dd, 2H, *J* = 11.7, 2.7 Hz), 4.30−4.42 (m, 4H), 7.23 (d, 2H, *J* = 7.8 Hz), 7.54−7.65 (m, 6H), 7.93 (dd, 2H, *J* = 7.8, 1.5 Hz), 8.17 (d, 2H, *J* = 7.8 Hz); ^{13}C NMR (CD_3OD) δ 19.3, 23.0, 26.2, 28.3, 30.4, 30.5, 30.7, 30.8, 35.2, 43.1, 53.1, 60.3, 64.3, 118.7, 123.2, 125.0, 125.1, 127.7, 128.3, 128.8, 133.1, 135.1, 147.7, 174.2, 180.3. Anal. ($C_{50}H_{64}N_2O_8{\cdot}0.75H_2O$) C, H, N.

Hexadecanenedioic acid bis[4-[(2*S*,3*R*,4*R*)-2-(hydroxymethyl)-4-isopropyl-5-oxo-3-pyrrolidinyl]-1-naphthalenyl] ester (4f): yield 90%; $[\alpha]_D$ +197 (*c* 0.9, MeOH); ^1H NMR (CD_3OD) δ 0.77 (d, 6H, *J* = 6.9 Hz), 0.83 (d, 6H, *J* = 6.9 Hz), 1.25−1.55 (m, 22H), 1.77−1.88 (m, 4H), 2.77 (t, 4H, *J* = 7.5 Hz), 2.96 (dd, 2H, *J* = 9.3, 3.9 Hz), 3.52 (dd, 2H, *J* = 11.4, 5.1 Hz), 3.77 (dd, 2H, *J* = 11.4, 2.7 Hz), 4.30−4.42 (m, 4H), 7.22 (d, 2H, *J* = 7.8 Hz), 7.53−7.65 (m, 6H), 7.93 (dd, 2H, *J* = 7.8, 1.5 Hz), 8.17 (d, 2H, *J* = 7.8 Hz); ^{13}C NMR (CD_3OD) δ 19.3, 23.0, 26.2, 28.3, 30.4, 30.5, 30.7, 30.8, 30.9, 35.2, 43.1, 53.1, 60.3, 64.3, 118.7, 123.2, 125.0, 125.1, 127.7, 128.3, 128.8, 133.1, 135.1, 147.7, 174.2, 180.3. Anal. ($C_{52}H_{68}N_2O_8{\cdot}0.5H_2O$) C, H, N.

Docosanedioic acid bis[4-[(2*S*,3*R*,4*R*)-2-(hydroxymethyl)-4-isopropyl-5-oxo-3-pyrrolidinyl]-1-naphthalenyl] ester (4g): yield 89%; $[\alpha]_D$ +164 (*c* 0.6, MeOH); ^1H NMR (CD_3OD) δ 0.77 (d, 6H, *J* = 6.9 Hz), 0.83 (d, 6H, *J* = 6.9 Hz), 1.22−1.55 (m, 34H), 1.77−1.88 (m, 4H), 2.77 (t, 4H, *J* = 7.5 Hz), 2.96 (dd, 2H, *J* = 9.3, 3.9 Hz), 3.52 (dd, 2H, *J* = 11.4, 5.1 Hz), 3.77 (dd, 2H, *J* = 11.4, 2.7 Hz), 4.30−4.42 (m, 4H), 7.22 (d, 2H, *J* = 7.8 Hz), 7.53−7.65 (m, 6H), 7.93 (dd, 2H, *J* = 8.1, 1.2 Hz), 8.17 (d, 2H, *J* = 8.1 Hz); ^{13}C NMR (CD_3OD) δ 19.3, 23.0, 26.3, 28.3, 30.4, 30.6, 30.7, 30.8, 30.9, 35.2, 43.1, 53.1, 60.3,

64.3, 118.7, 123.2, 125.0, 125.1, 127.7, 128.3, 128.8, 133.1, 135.1, 147.7, 174.2, 180.3. Anal. ($C_{58}H_{80}N_2O_8 \cdot 0.5H_2O$) C, H, N.

9-[*N*-(4-Acetoxy-1-naphthalenyl)carbamoyl]nonanoic Acid(6). 4-Hydroxy-1-naphthylamine hydrochloride (0.45 g, 2.2 mmol) was taken up in dry tetrahydrofuran (5 mL) and cooled to 0 °C, and triethylamine (0.65 mL, 4.4 mmol) was added. Then methyl 9-(chlorocarbonyl)nonanoate[41] (0.5 g, 2.2 mmol) in dry tetrahydrofuran (2.5 mL) was added slowly, and the reaction mixture was stirred at room temperature overnight. The mixture was diluted with ethyl acetate, washed with water and brine, and dried over anhydrous sodium sulfate. After evaporation, the residue was purified on silica gel (2% methanol in chloroform) to obtain the amide, which was dissolved in methanol (5 mL). Lithium hydroxide (100 mg) was added, and the mixture was stirred at room temperature for 0.5 h. Acidification with 6 N HCl and extraction with ethyl acetate provided the free acid (0.21 g, 28% yield). To a solution of this acid in dichloromethane was added potassium carbonate (80 mg) and acetic anhydride (0.3 mL). The mixture was stirred at room temperature overnight. Water (5 mL) was added, and the mixture stirred for another 1 h. The dichloromethane layer was separated. The water layer was extracted twice more with dichloromethane and the combined extracts were concentrated to obtain the acetate (0.20 g, 91% yield): ^{1}H NMR (CD_3OD) δ 1.23−1.48 (m, 8H), 1.52−1.68 (m, 2H), 1.70−1.79 (m, 2H), 2.27 (t, 2H, J = 7.2 Hz), 2.41−2.51 (m, 5H), 7.20 (d, 1H, J = 7.8 Hz), 7.34 (br s, 1H), 7.46−7.55 (m, 1H), 7.62 (d, 1H, J = 7.8 Hz), 7.86 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 24.9, 25.8, 29.00, 29.05, 29.07, 29.2, 341, 37.0, 51.5, 108.4, 121.2, 122.8, 123.5, 124.7, 125.3, 126.5, 130.1, 151.7, 174.1, 174.2.

9-[*N*-(4-Hydroxy-1-naphthyl)carbamoyl]nonanoic Acid *N*-[(2*S*,5*S*)-1,2,3,4,5,6-hexahydro-5-(hydroxymethyl)-2-isopropyl-1-methyl-3-oxobenzo[*e*][1,4]diazocin-8-yl]-amide (7). The nitro compound **5** (50 mg, 0.144 mmol) was dissolved in ethanol (8 mL), and 10% Pd/C (7.5 mg, 0.007 mmol) was added. The mixture was stirred vigorously under an H_2 atmosphere overnight. The reaction mixture was filtered and the filtrate was concentrated to give a residue that was dried under vacuum for 1 h at 60 °C. To a solution of the residue in dimethylformamide was added successively the acid **6** (37 mg, 0.12 mmol) and 1-[3′-(dimethylamino)propyl]-3-ethyl carbodiimide hydrochloride (22 mg, 0.12 mmol). The mixture was stirred at room temperature for 12 h. Water was added, and the mixture was extracted with dichloromethane (thrice). The combined organic phases were dried over anhydrous sodium sulfate and concentrated. The residue was dissolved in ethanol (30 mL). A solution of sodium carbonate (60 mg) in water (6 mL) was added with vigorous stirring. After 1.5 h at room temperature, the solution was concentrated and an additional amount of water (20 mL) was added. Extraction with ethyl acetate (4 × 30 mL) followed by preparative TLC (ethyl acetate as developing solvent) provided the product (23 mg, 35% yield): $[\alpha]^{20}_D$ −75.2 (*c* 0.35, CH_3OH); ^{1}H NMR (CD_3OD) δ 0.95 (d, 3H, J = 6.8 Hz), 1.08 (d, 3H, J = 6.8 Hz), 1.25−1.29 (m, 8H), 1.68−1.82 (m, 4H), 2.01−2.08 (m, 1H), 2.29−2.52 (m, 4H), 2.72 (s, 3H), 2.88−2.94 (m, 2H), 3.35−3.39 (m, 1H), 3.46−3.52 (m, 1H), 3.61−3.68 (m, 2H), 4.51 (br s, 1 H), 6.79−6.85 (m, 1H), 7.04−7.07 (m, 1H), 7.29−7.33 (m, 2H), 7.44−7.53 (m, 3H), 7.82 (d, 1H, J = 7.2 Hz), 8.25 (d, 1H, 7.2 Hz); ^{13}C NMR ($CDCl_3$) δ 18.9, 20.1, 25.5, 25.8, 28.8, 28.9, 29.6, 37.3, 47.6, 48.1, 48.4, 48.7, 48.9, 49.3, 56.6, 64.3, 87.0, 102.5, 107.3, 119.4, 119.7, 121.6, 121.74, 124.2, 124.7, 126.4, 147.8, 174.9, 176.2, 186.2; HPLC retention time 6.8 min (99.96% purity) using conditions C; FABMS calcd for ($C_{35}H_{46}N_4O_5$ + H^+) 603.36, found 603.37.

Acknowledgment. We are indebted to the National Institutes of Health (Grant CA79601) for support of this work. We thank Dr. Werner Tückmantel for proofreading the manuscript.

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JM0302041