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3-Hydroxy-3-Methylglutaryl-Coenzyme A Reductase Inhibitors: Oxime Ether Analogs of Pravastatin

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Abstract—Pravastatin, a potent anti-hypercholesteremic drug, has been developed by Bristol-Myers Squibb for treatment of hypercholesterolemia and other related diseases. Several structurally related compounds (SQ 31,554, SQ 31,879, SQ 31,947, SQ 32,391, SQ 32,770, SQ 32,390 and SQ 32,469) modified at the 3-position of the hexahydronaphthalene ring system of pravastatin were prepared in the course of developing the basic reagents for a radioimmunoassay of the parent drug. The biological activity of these analogs was comparable to pravastatin itself. Indeed, one member of this series was found to be several times more potent than pravastatin.

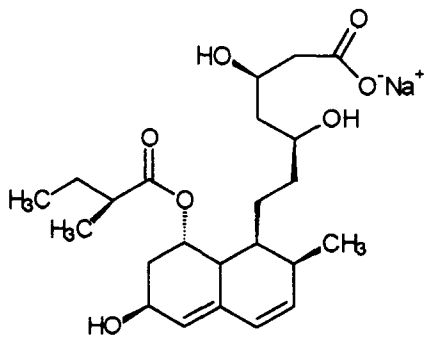
Introduction

Pravastatin (**I**) is a potent anti-hypercholesteremic drug belonging to the same family of HMG-CoA reductase inhibitors as lovastatin (**II**) and simvastatin (**III**). HMG-CoA reductase is a key enzyme in the biosynthesis of endogenous cholesterol in humans.[1] Excessive levels of serum cholesterol and LDL-conjugated cholesterol have been implicated in the development of atherosclerosis and coronary artery disease.[2] On this basis, compounds like pravastatin represent an active area of investigation in the pharmaceutical industry.[3]

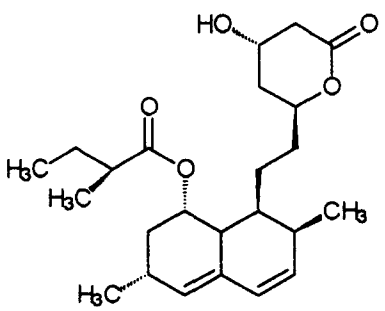
In the course of developing a radioimmunoassay for pravastatin, several modifications to the parent drug were required to prepare reagents for subsequent immuno-stimulation and radiotracer development. It was found convenient to restrict these modifications to the 3-position of the hexahydronaphthalene ring system of pravastatin. As several structurally related compounds modified in the 3-position were shown to retain the activity of the parent drug,[4,5] we set out to prepare a representative series of compounds with relevance as radioimmunoassay reagents. Overall, we were surprised at the degree to which these analogs retained the activity of the parent drug. Indeed, one member of the series was found to be several times more potent than pravastatin. We report herein the syntheses of these derivatives and their inhibitory activity with respect to HMG-CoA reductase.[6]

Chemistry

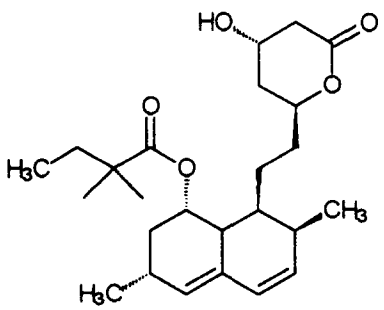
Our initial attempts at preparing suitable radioimmunoassay reagents for pravastatin involved activation and subsequent amidation of the terminal carboxylic acid. Unfortunately, all attempts at activation of this moiety resulted in prompt γ-lactone



PRAVASTATIN (I)



LOVASTATIN (II)



SIMVASTATIN (III)

Figure 1.

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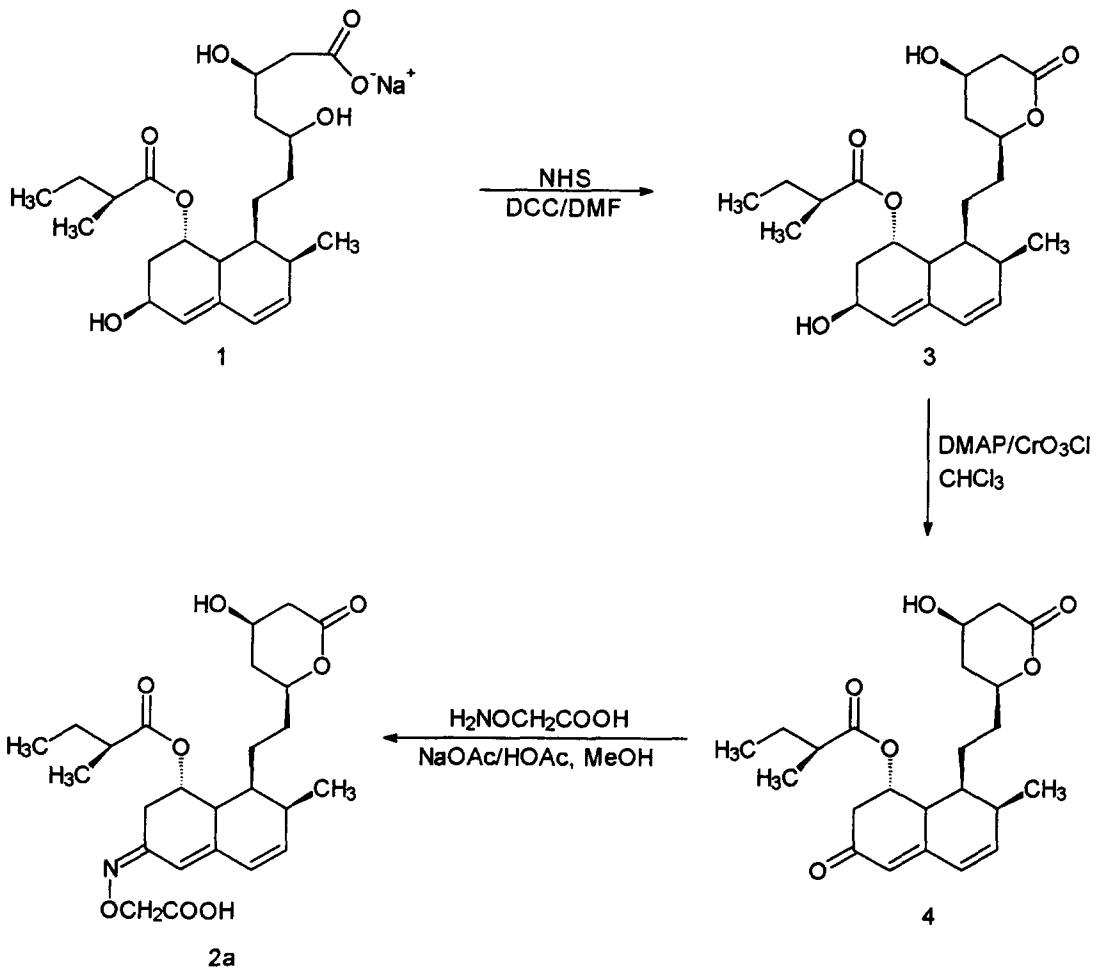
formation, which in turn proved to be a poor acylating agent. In fact, simvastatin and lovastatin are administered as their lactones, although it is believed that the hydroxyacid is the active species and that the lactone is in equilibrium with the acid *in vivo*.[7] A potential loss of biological activity upon amidation of the terminal carboxylic acid was also a cause for concern. Therefore, derivatization of the decalin ring system, specifically at the 3-hydroxy position, was considered to be a reasonable strategy. Thus, oxidation of the allylically-activated hydroxyl group at the 3 position of the decalin ring system and conversion to its carboxymethyl oxime-ether provides a free carboxylic acid which could then be conjugated to radioiodinatable moieties such as histamine or phenol derivatives. Also, the cold iodinated histamine conjugate and iodinated phenol derivatives would be prepared as standards and evaluated for their HMG-CoA reductase inhibitory activity.

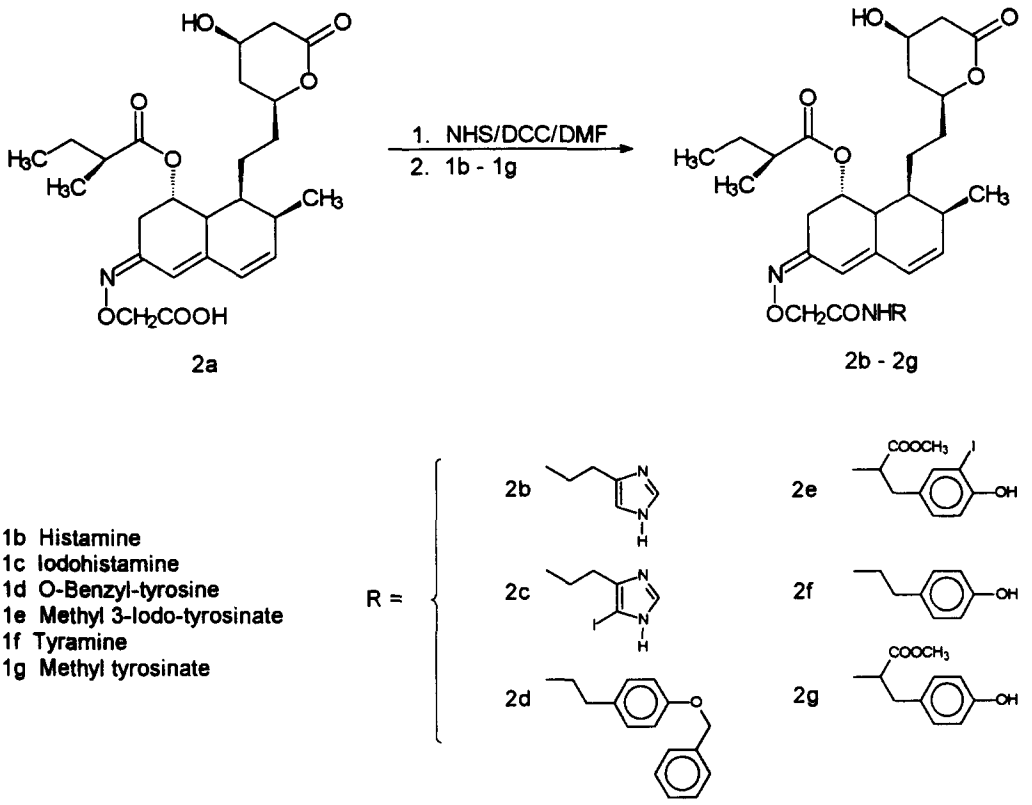
As mentioned above, the activation of the side chain carboxylate resulted in lactonization. The lactone formation not only improves the organic solubility and simplifies purification, it also serves to mask the hydroxyacid. Thus, treatment of pravastatin with DCC/*N*-hydroxysuccinimide in DMF resulted in good yield (71%) of lactone. Selective oxidation of the diallylic 3-β-hydroxyl was attempted by several methods, including activated MnO_2, both the commercial material[8] and that prepared in the laboratory,[9] dimethylpyrazole/PCC[10] and DMAP/CrO_3Cl.[11] Of these methods, the latter proved to be the best in terms of yield and ease of purification. Thus, treatment of the lactone of pravastatin with eight equivalents of DMAP/CrO_3Cl in $CHCl_3$ at room temperature afforded the dienone in moderate (27%) yield. This material proved to be photo-labile as well as heat-sensitive.

Treatment of the dienone with carboxymethoxylamine in anhydrous methanol provided the oxime ether in good (55%) yield.[12] This material was not as sensitive as the dienone with regard to ambient exposure (Scheme 1).

Subsequent activation of the carboxylic group of the oxime ether with *N*-hydroxysuccinimide/DCC in DMF solution followed by treatment with histamine, iodohistamine, *O*-benzyl-tyramine, methyl 3-iodotyrosinate, tyramine and methyl tyrosinate in phosphate buffer (pH 7.4) afforded **2b**, **2c**, **2d**, **2e**, **2f** and **2g**, respectively, in moderate to good yield.



Scheme 1.

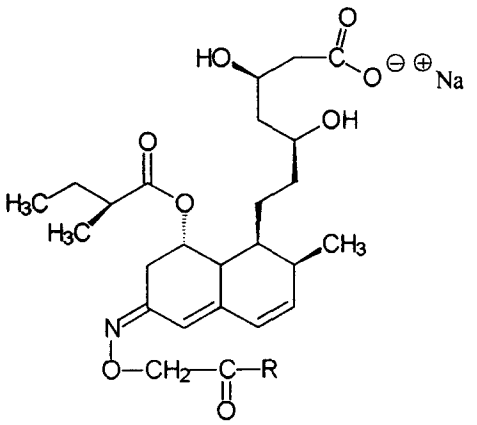


Scheme 2.

Biological Results and Discussion

The oxime ethers listed in Table 1 were submitted for testing as lactones or were converted to their respective ring-opened dihydroxy acid sodium salts prior to submission and evaluated for their intrinsic inhibitory activity against HMG-CoA reductase.[14] In addition, all analogs were evaluated as mixtures of *syn*- and *anti*-isomers, since rapid equilibration of these isomers was observed at physiological pH.

Table 1. *In vitro* HMG-CoA reductase inhibitory activity



Compound	No.	R	IC_{50}, nM	Relative potency
Pravastatin	1	—	26.0	1.0
SQ 31,554	2a	OH	206.0	0.13
SQ 31,879	2b	Histamine	37.0	0.70
SQ 31,947	2c	Iodohistamine	9.3	2.80
SQ 32,391	2d	*O*-Benzyltyramine	91.2	0.29
SQ 32,770	2e	Methyl 3-iodo-tyrosinate	210.2	0.12
SQ 32,390	2f	Tyramine	38.4	0.67
SQ 32,469	2g	Methyl tyrosinate	100.5	0.26

Despite the marked structural similarity of pravastatin, lovastatin and simvastatin, minor variations in the decalin ring have resulted in major differences in their physicochemical and pharmacokinetic properties. The active inhibitory species of these compounds are the hydroxyacids, present as such in pravastatin and produced as a result of lactone hydrolysis in the other two drugs. The hydroxyacid side chain of the drug occupies the hydroxymethylglutaryl binding site on the enzyme. However, mevalonic acid, which is structurally analogous to the hydroxyacid side chain, is not a potent inhibitor of HMG-CoA reductase. It can therefore be concluded that the decalin ring system in these drugs must reside in a hydrophobic region of the enzyme, thus serving to direct and anchor the side chain at the active site[13] and rendering the drugs potent inhibitors of HMG-CoA reductase. The hydrophobic pocket can accommodate a variety of compounds with different structures, suggesting that there is a less stringent structural constraint there than in the interaction between the enzyme active site and the substrate. It also suggests that the hydrophobic interaction is a result of a number of interactions of lower affinity rather than a single interaction of high affinity.[7] Further, it is possible that the polar groups resident in the amide side chains of the pravastatin analogs we have synthesized have added another dimension through new hydrogen bond interactions between the various molecules and the hydrophobic region of the enzyme, thus increasing the affinity of these compounds for the enzyme.

In summary, the oxime ether analogs of pravastatin possessed a surprising degree of inhibitory activity to HMG-CoA reductase. Of the compounds tested, compound **2c** was the most potent.

Experimental

General methods

Pravastatin was provided by the Bristol-Myers Squibb Pharmaceutical Research Institute. Iodohistamine was prepared by a modification of the method by Tantchou and Slaunwhite.[15] *O*-Benzyl tyramine was obtained from the Bader Library of Rare Chemicals, Division of Aldrich, Inc. It can also be prepared by the reduction of the corresponding nitro-styrene.[16]

Unless otherwise stated, all other compounds were obtained from Aldrich and were used without further purification. Proton NMR spectra were recorded in $CDCl_3$ on a JEOL (Japan, Electron Optics, Ltd) 270 or 400 MHz spectrometer; chemical shifts are reported in ppm downfield from Me_4Si as the internal standard. Mass spectra were taken on either a Finnigan TSQ or a FAB VG-ZAB-2F mass spectrometer. Thin layer chromatography (TLC) was performed with precoated silica gel 60F-254 plates from E.M. Science. High performance liquid chromatography (HPLC) was done using a Spectra-physics chromatographic unit. Column chromatography was carried out using Kieselgel 60 (70–230 mesh) from E.M. Science. Elemental analyses were performed by the Microanalytical Laboratory of the Bristol-Myers Squibb Pharmaceutical Research Institute and are within 0.4% of the theoretical value.

Biology

Compounds **2a** and **2b** were submitted for testing as their sodium carboxylates. All the other compounds were submitted as lactones but were hydrolyzed prior to analysis. The relative potency of each compound was determined by comparing its IC_{50} with that of pravastatin, which was tested and arbitrarily assigned a relative potency of 1.

Rat hepatic HMG-CoA reductase activity was measured using a modification of the method described by Edwards.[14] Rat hepatic microsomes were used as a source of the enzyme and the enzyme activity was determined by measuring the conversion of the ^{14}C-HMG-CoA substrate to ^{14}C mevalonic acid.

Chemistry

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-3-hydroxy-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 31,369) (3).* Pravastatin (3.32 g, 7.44 mmol), dicyclohexylcarbodiimide (3.47 g, 16.8 mmol) and *N*-hydroxysuccinimide (1.93 g, 16.8 mmol) were stirred in 50 mL of dry DMF under nitrogen for 16 h. The resulting suspension was filtered and the filtrate evaporated at 30 °C under reduced pressure. The residue was taken up in 50 mL of 20:1 $CHCl_3$:MeOH, filtered and evaporated. The resulting yellow oil was purified by column chromatography on silica gel using a gradient solvent system consisting of $CHCl_3$:MeOH from 1 to 10% CH_3OH. The title compound was eluted with 20:1 $CHCl_3$:MeOH, evaporated to dryness and triturated with ether to yield a white powder (2.15 g, 71%). HPLC: (65% H_2O:CH_3OH (9:1) + 35% CH_3CN, Alltech Lichrosorb RP-18, 5 μ, 25 cm column, 1.0 mL min^{-1}, UV 250 nm); R_t 15.28 min; MS: $(M+H)^+$ 407, IR (cm^{-1}): 3501, 2904, 2935, 1727, 1079, 1017; ^1H NMR: δ 0.89 (overlapping *d* and *t*, 6H), 1.12 (*d*, 3H, *J* = 7.0 Hz), 1.2–2.1 (complex, 12H), 2.35 (*m*, 3H), 2.5–2.68 (*m* and *dd*, 2H), 2.74 (*dd*, 1H, *J* = 5,18 Hz), 4.37 (*br s*, 1H), 4.62 (*m*, 1H), 5.4 (*m*, 1H), 5.61 (*s*, 1H), 5.91 (*dd*, 1H, *J* = 6.0, 9.6 Hz), 6.05 (*d*, 1H, *J* = 9.7 Hz). Anal.($C_{23}H_{34}O_6$): C,H,O.

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-7-methyl-3-oxo-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 31,553) (4).* The lactone SQ 31,369 (1.0 g, 2.33 mmol) and CrO_3Cl-dimethylaminopyridine (DMAP) (5.1 g, 19.7 mmol) were stirred in 250 mL of $CHCl_3$ under nitrogen in the dark for 18 h. The dark suspension was then filtered over a bed of Celite and the filtrate evaporated at ambient temperature under reduced pressure. The dark residue was chromatographed over silica gel eluting with $CHCl_3$:IPA (9:1), evaporated to

dryness and triturated with pentane to yield the title compound, the dienone, as a white powder (255 mg, 27%). HPLC: (65% H_2O:CH_3OH (9:1) + 35% CH_3CN, Alltech Lichrosorb RP-18, 5 μ, 25 cm column, 1.0 mL min^{-1},UV 250 nm); R_t 18.8 min; MS: $(M+H)^+$ 405; IR (cm^{-1}): 3203, 3132, 3110, 3040, 1726, 1596, 1485. Anal. $C_{23}H_{32}O_6.0.5H_2O$: C,H,O.

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]]-2-Methylbutanoic acid,3-[(carboxymethoxy)imino]-1,2,3,7,8,8a-hexahydro-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 31,554) (2a).* The dienone SQ 31,553 (178 mg, 0.44 mmol) was stirred with carboxymethoxylamine hemihydrochloride (340 mg, 1.54 mmol) and anhydrous sodium acetate (133 mg, 1.62 mmol) in 30 mL of absolute CH_3OH containing 15 drops of glacial HOAc for 18 h at ambient temperature. The reaction mixture was diluted with 125 mL of a mixture of $CHCl_3$:MeOH (5:1) and washed with water (4 × 40 mL). The organic layer was dried, filtered and evaporated under reduced pressure to an oil, which was chromatographed on silica gel eluting with $CHCl_3$:IPA:HOAc (6:1:0.2). The resulting oil was co-evaporated with 2 volumes of cyclohexane to yield the title compound, **2a** (116 mg, 55%). HPLC: (50% H_3PO_4 (0.1%, pH 5) containing 1% KCl, + 25% CH_3OH + 25% CH_3CN, Alltech Lichrosorb RP-18, 5 μ, 25 cm column, 1.0 mL min^{-1}, UV 250 nm); R_t 7.34 and 8.97 min; MS: $(M+H)^+$ 478, $(M+H)^-$ 477; IR (cm^{-1}): 3202, 2926, 2878, 1724, 1596, 1467, 1307, 1029. Anal. $C_{25}H_{35}O_8 \cdot H_2O$: C,H,N.

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-3-[[2-[[2-(1H-imidazole-4-yl)ethyl]amino]-2-oxoethoxy]imino]-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 31,879) (2b).* A mixture of **2a** (100 mg, 0.21 mmol), *N*-hydroxysuccinimide (54.5 mg, 0.47 mmol) and dicyclohexylcarbodiimide (97.6 mg, 0.47 mmol) in 6 mL of dry DMF was stirred under nitrogen for 2 h, at which time histamine (69.8 mg, 0.63 mmol) in 2 mL of potassium phosphate buffer (pH 7.5) was added in one portion. The mixture was stirred for 2 h. The reaction mixture was taken up in $CHCl_3$:MeOH (3:1), filtered, concentrated and chromatographed over silica gel eluting with $CHCl_3$:MeOH (3:1) to yield the title compound, **2b** (60 mg, 50%). HPLC: (50% H_3PO_4. (0.1%, pH 5) containing 1% KCl, + 25% CH_3OH + 25% CH_3CN, Alltech Lichrosorb RP-18, 5 μ, 25 cm column, 1.0 mL min^{-1}, UV 250 nm); R_t18.08 and 23.3 min; MS: $(M+H)^+$ 571, $(M+H)^-$ 569; IR (cm^{-1}): 3404, 2963, 1726, 1563, 1554, 1435, 1081. ^1H NMR: δ 0.89 (overlapping *d* and *t*, 6H), 1.12 (*d*, 3H, *J* = 7.0 Hz), 1.3–2.1 (complex, 12H), 2.3–2.85 (complex, 8H), 3.5 (complex, 2H), 4.2 (*m*, 1H), 4.45 (*d*, 2H), 4.7 (*m*, 1H), 5.45 (complex, 1H), 5.95 (complex, 1H), 6.15 (complex, 2H), 6.6 (*s*, 1H), 6.9 (*s*, 1H), 7.65 (*s*, 1H). Anal. $C_{30}H_{42}N_4O_7 \cdot 2H_2O$: C,H,N.

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-3-[[2-[[2-(5-iodo-1H-imidazole-4-yl)ethyl]amino]-2-oxoethoxy]imino]-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 31,947) (2c).* A mixture of **2a** (100 mg, 0.21 mmol), *N*-hydroxysuccinimide (54.5 mg, 0.47 mmol) and dicyclohexylcarbodiimide (97.6 mg, 0.47 mmol) in 6 mL of dry DMF was stirred in a dry atmosphere for 2 h, at which time 5-iodohistamine dihydrochloride[15] (91 mg, 0.29 mmol) in 3 mL of potassium phosphate buffer (pH 7.4) was added in one portion. The mixture was stirred overnight. The resulting suspension was filtered, concentrated and the residue chromatographed over silica gel eluting with $CHCl_3$: MeOH (9:1) to yield the title compound, **2c** (78 mg, 53%). HPLC: (46% H_3PO_4, 0.1%, pH 5) containing 1% KCl, + 27% CH_3OH + 27% CH_3CN, Alltech Lichrosorb RP-18, 5 μ, 25 cm column, 1.0 mL min^{-1}, UV 250 nm); R_t 24.4 and 30.8 min; MS: $(M+H)^+$ 697, $(M+H)^-$ 696 IR (cm^{-1}): 3408, 2964, 1725, 1078; ^1H NMR: δ 0.89 (overlapping *d* and *t*, 6H), 1.12 (*d*, 3H, *J* = 7.0 Hz), 1.3–2.1 (complex, 12H), 2.32 (*m*, 2H), 2.52 (*m*, 2H), 2.7 (*m*, 2H), 2.85 (*m*, 2H), 3.50 (complex, 2H), 4.4 (*m*, 1H), 4.45 (*d*, 2H), 4.65 (*m*, 1H), 5.45 (*br s*, 1H), 6.15 (complex, 2H), 6.6 (*s*, 1H), 7.55 (*s*, 1H). Anal. $C_{30}H_{41}IN_4O_7 \cdot 3H_2O$: C,H,N.

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-3-[[2-[[2-(4-phenylmethoxy)-phenyl]ethyl]amino]-2-oxoethoxy-imino]-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 32,391) (2d).* A mixture of **2a** (50 mg, 0.11 mmol), *N*-hydroxysuccinimide (30.2 mg, 0.26 mmol) and dicyclohexylcarbodiimide (52.0 mg, 0.25 mmol) in 10 mL of dry DMF was stirred under nitrogen for 2.5 h at which time *O*-benzyltyramine (72 mg, 0.31 mmol) was added in one portion with stirring. The reaction mixture was stirred overnight at ambient temperature, filtered, concentrated and chromatographed by reverse phase preparative HPLC: (buffer H_3PO_4. (0.1%, pH 5) containing 1% KCl + CH_3OH + CH_3CN, 0–15 min 40:30:30, 15–40 min 25:38:37, Dynamax C_{18}, flow rate 20 mL min^{-1}; R_t 30.1 and 31.7 min to yield the title compound, **2d** (38 mg, 53%) as a white amorphous powder. TLC: $CHCl_3$:CH_3OH (10:1) R_f 0.68. Anal. $C_{40}H_{50}N_2O_8$: C,H,N.

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-3-[[2-[[1-(4-hydroxy-3-iodophenyl)methyl]-2-methoxy-2-oxoethyl]amino]-2-oxoethoxy-imino]-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 32,770) (2e).* A mixture of **2a** (50 mg, 0.11 mmol), *N*-hydroxysuccinimide (27.2 mg, 0.24 mmol) and dicyclohexylcarbodiimide (48.8 mg, 0.24 mmol) in 6 mL of dry DMF was stirred under nitrogen for 2.5 h, at which time methyl 3-iodo-tyrosinate hydrochloride (112 mg, 0.31 mmol) in 3 mL of DMF containing 50 μL of triethylamine was added in one portion. The reaction mixture was stirred overnight at ambient temperature. The resulting suspension was filtered, concentrated and chromatographed on silica gel eluting with $CHCl_3$: MeOH: HOAc (10:1:0.1) to yield the title compound, **2e** (30.7 mg, 36%) as a white amorphous powder. MS: $(M+H)^+$ 781, $(M+H)^-$ 779; IR (cm^{-1}): 3424, 3009, 1728,

1671, 1507, 1489, 1078. Anal $C_{35}H_{45}IN_2O_{10}\cdot 2.75H_2O$: C,H,N.

[1S-[1α(R),7β,8χ(2S*,4S*),8aβ]]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-3-[[2-[[2-(4-hydroxyphenyl)-ethyl]amino]-2-oxoethoxy]imino]-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 32,390) (2f).* A mixture of **2a** (50 mg, 0.11 mmol), *N*-hydroxysuccinimide (27.2 mg, 0.24 mmol) and dicyclohexylcarbodiimide (48.8 mg, 0.24 mmol) in 6 mL of dry DMF was stirred under nitrogen for 2.5 h, at which time tyramine (43.2 mg, 0.31 mmol) in a mixture of 4 mL of DMF and 1.5 mL of potassium phosphate buffer (pH 7.4) was added in one portion with stirring. The mixture was stirred for 3 h at ambient temperature, filtered, concentrated and chromatographed over silica gel eluting with $CHCl_3$:MeOH (9:1) to yield the title compound, **2f** (25 mg, 38%) as an amorphous solid. ^{1}H NMR: δ 0.82 (*t*, 3H), 0.99 (*d*, 3H, *J* = 7.0 Hz), 1.13 (*d*, 3H, *J* = 7.0 Hz), 1.3–2.1 (complex, 12H), 2.3 (*m*, 2H), 2.5 (*m*, 2H), 2.62 (*m*, 4H), 3.4 (complex, 2H), 4.22 (*s*, 1H), 4.42 (*d*, 2H), 4.65 (*m*, 1H), 5.42 (*br s*, 1H), 5.95 (*br s*, 1H), 6.2 (*d*, 2H), 6.65 (*dd*, 2H), 7.0 (*dd*, 2H). Anal. $C_{33}H_{44}N_2O_8\cdot H_2O$: C,H,N.

[1S-[1α(R),7β,8β(2S*,4S*),8aβ]]-2-Methylbutanoic acid, 1,2,3,7,8,8a-hexahydro-3-[[2-[[1-(4-hydroxyphenyl)methyl]-2-methoxy-2-oxoethyl]amino]-2-oxoethoxyimino]-7-methyl-8-[2-(tetrahydro-4-hydroxy-6-oxo-2H-pyran-2-yl)ethyl]-1-naphthalenyl ester (SQ 32,469) (2g).* A mixture of **2a** (50 mg, 0.11 mmol), *N*-hydroxysuccinimide (27.2 mg, 0.24 mmol) and dicyclohexylcarbodiimide (48.8 mg, 0.24 mmol) in 6 mL of dry DMF was stirred under nitrogen for 2.5 h, at which time methyl tyrosinate (72.8 mg, 0.31 mmol) in 0.5 mL of DMF was added in one portion with stirring. The mixture was stirred for 3 h at ambient temperature, filtered, concentrated and chromatographed over silica gel eluting with $CHCl_3$:MeOH (10:1) to yield the title compound, **2g** (27 mg, 38%) as an amorphous solid. MS: $(M+H)^+$ 655, $(M+H)^-$ 654; IR (cm^{-1}): 3434, 2924, 1728, 1686, 1029; ^{1}H NMR: δ 0.82 (*t*, 3H), 0.99 (*d*, 3H, *J* = 7.0 Hz), 1.13 (*d*, 3H, *J* = 7.0 Hz), 1.3–2.1 (complex, 12H), 2.3 (*m*, 2H), 2.6 (complex, 2H), 2.75 (*s*, 1H), 2.90 (*s*, 2H), 3.6 (*d*, 3H), 4.1 (*s*, 1H), 4.4 (*m*, 3H), 5.15 (*d*, 1H), 5.3 (*d*, 1H), 5.95 (*br s*, 1H), 6.2 (complex, 2H), 6.6 (*dd*, 2H), 7.0 (*dd*, 2H), 7.82 (*d*, 1H), 9.15 (*s*, 1H). Anal. $C_{35}H_{46}N_2O_{10}\cdot 0.5H_2O$: C,H,N.

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