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3,4-Disubstituted oxazolidin-2-ones as constrained ceramide analogs with anticancer activities

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ABSTRACT

Heterocyclic analogs of ceramide as 3-alkanoyl or benzoyl-4-(1-hydroxy-2-enyl)-oxazolidin-2-ones were designed by binding of primary alcohol and amide in sphinogosine backbone as a carbamate. They were synthesized by addition of acyl halide to the common ring 4-(1-*t*-butyldimethylsilyloxyhexadec-2-enyl)-oxazolidin-2-one which was elaborated from chiral aziridine-2-carboxylate including stereoselective reduction and ring opening reactions as key steps. Other analogs with different carbon frame at C4 position which is corresponding to the sphingoid backbone were prepared from 3-cyclopentanecarbonyl-4-(1-*t*-butyldimethylsilyloxybut-2-enyl)-oxazolidin-2-one and straight and cyclic alkenes by cross metathesis. All compounds were tested as antileukemic drugs against human leukemia HL-60 cells. Many of them including propionyl, cyclopentanoyl and *p*-nitrobenzoyl-4-(1-hydroxyhexadec-2-enyl)-oxazolidin-2-ones showed better antileukemic activities than natural C2-ceramide with good correlation between cell death and DNA fragmentation. There is a drastic change of the activities by the carbon chain lengths at C4 position. Cytotoxicity was induced by caspase activation without significant accumulation of endogenous ceramide concentration or any perturbation of ceramide metabolism.

1. Introduction

Sphingolipids including ceramide, sphingomyelin, cerebroside, and ganglioside are unique membrane components of all eukaryotic cells.[1] They play important roles in the regulation of cell proliferation, differentiation, migration, survival, and cell-death via closely related metabolic pathways.[1,2] Among all metabolites ceramide is one of the most important molecules as a secondary messenger of sphingolipid signaling pathway.[2]

The main structural components of ceramide (**1**) are a long alkyl chain with 2-amino-1,3-diols and C4,5-*trans*-olefin as the sphingosine backbone and an *N*-acyl group. In mammalian tissue the most common sphingoid base is D-*erythro*-C18-sphingosine [(2*S*,3*R*,4*E*)-2-amino-octadec-4-en-1,3-diol, (*E*)-sphing-4-enine] and the typical acyl group attached on the amine has the chain length of 16–24. The structural changes of ceramide at the primary alcohol occur at the cellular metabolic system by addition of phosphate, phosphocholine and carbohydrates to form ceramide-1-phosphate (**2**), sphingomyelin (**3**) and glucosylceramide (**4**) (Fig. 1). Specifically ceramide is generated from sphingosine by ceramide synthase with an additional fatty acid chain to the sphingoid backbone. Hydrolysis of sphingomyelin bearing additional phosphocholine to ceramide is also important pathway for ceramide[3] as a secondary messenger by sphingomyelinase[4] in response to extracellular agents and stresses including chemotherapeutic agents[5] tumor necrosis factor α (TNF-α),[6] and ionizing radiation.[7] The dynamic balance of the sphingolipid metabolites is an important factor whether a cell undergoes proliferation or programmed death so called apoptosis.[8] The close relationship is found between ceramide and its hydrolytic products sphingosine triggered by growth factors such as platelet-driven growth factors. Ceramide as an important stimulating factor of apoptosis is hydrolyzed by ceramidase to sphingosine followed by activation of kinase to yield sphingosine-1-phosphate with cell growth.

Lots of sphingosine analogs were isolated from natural sources and synthesized based on the structural modification of sphingoid backbones, and their biological activities were evaluated. Among them the synthetic analog safingol and the marine natural product spisulosin were good examples. Safingol is an inhibitor of protein kinase C[9] and spisulosin inhibits the activity of Rho protein.[10] Few years ago we envisaged the synthetic method for the construction of sphingoid backbone and their analogs including anticancer agent's spisulosin and safingol based on the stereospecific reduction of 2-acylaziridine and aziridine ring opening reactions.[11]

Among the enormous increase in the research of sphingolipid metabolism the central metabolite ceramide attracts most attentions due to its important role at the stages of apoptosis with a

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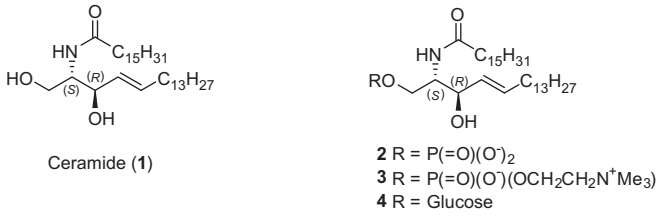


Figure 1. Structures of ceramide (**1**) ceramide-1-phospahate (**2**), sphingomyelin (**3**) and glucosylceramide (**4**).

selective toxicity to malignant but not to normal cells.[12] Various analytical methods show that ceramide is accumulated during the initiation stage of apoptosis.[3] The central molecule in the sphingolipid metabolism, ceramide, is known to induce apoptosis, senescence,[13] and growth arrest[14] in many human cancers. Therefore, many researchers have focused on designing ceramide analogs with the same apoptotic activity as ceramide. They lead synthesizing dozens of active molecules reported recently, many of which have greater inhibitory effect on the enzymes involved in ceramide metabolism.[15]

Exogenous ceramide actually induces apoptotic programmed cell death. In particular, cell-permeable ceramide with short acyl group such as C2-Cer (*N*-acetyl-D-*erythro*-C18-sphingosine) and C8-Cer (*N*-octyl-D-*erythro*-C18-sphingosine) inhibit tumor cell growth and induce apoptosis in leukemia cell lines.[16] The ceramides with longer alkyl chain than C8-Cer including natural ceramide (C18-Cer) are too lipophilic to pass through the cell membrane. Dihydroceramide (DHCer) with saturated D-*erythro*-C18-sphinganine does not have any biological activity which seems to be due to the lack of essential *trans* allylic alcohol.[17] For this reason, many synthetic and natural ceramide analogs were prepared and evaluated as anticancer agents by altering the backbone and by changing two hydroxy and one amine functional groups. Those include ceramide analogs whose backbone was modified with various aromatic or heteroaromatic systems instead of simple long alkyl chain of sphingoid base with free hydroxyl group at the end of the backbone.[18] However, most of them were less active than C2-Cer. Furthermore, acyclic ceramide analogs bearing primary alcohol in sphingoid backbone perturb or at least have a chance to perturb the metabolic pathways by its possible involvement in various biological reactions.

Constrained ceramide analogs without the primary alcohol may overcome this drawback of metabolic perturbation. A good example was shown by uracil ring compound in which the polar portion of ceramide was replaced.[19] This compound has very potent growth-inhibitory effect of CCRF-CEM human leukemia cells to trigger apoptosis by the occurrence of DNA and nuclear fragmentation.[19] However, these uracil analogs do not bear an allylic alcohol system and do have aromaticity in the main ring part, which are quite different from the real conformational alignment of acyclic C2-Cer.

Recently we reported a short communication for the synthesis and biological activities of new constrained ceramide analogs which are closer to the real conformational and electronic properties of ceramide on the basis of oxazolidin-2-one ring (**5**).[20] Analogs with oxazolidin-2-one ring were designed to combine the primary alcohol at C1 and the amide nitrogen at C2 of ceramide together by additional carbonyl group. This conformation was deduced by early observation that there are strong hydrogen-bonding among alcoholic hydrogens and the amide nitrogen.[21] An analog, *N*-hexadecanoyl compound **5c**, mimicked naturally occurring acyclic C16-Cer whose conformational similarity was evaluated by the comparison with the minimum energy conformers calculated by density functional model.[20] The overlay structure of those two compounds, C16-Cer and **5c**, shows similarity in Figure 2.

This analog retains the same conformation with the allylic alcohol system as for the natural substrate that is one of the essential structural elements to keep the biological activity with the generation of reactive oxygen species.[2c,17b] Furthermore, in this analog the free primary hydroxyl group of the natural substrate was bound to the oxazolidin-2-one ring that may block metabolic perturbation occurring at the hydroxyl group of C-1 of the sphingoid backbone. Thereby we decided to prepare 3-alkanoyl or benzoyl-4-(1-hydroxy-2-enyl)-oxazolidin-2-ones as heterocyclic analogs of ceramide and to test for antileukemic drugs against human leukemia HL-60 cells.

2. Results and discussion

All of the designed compounds were synthesized from commercially available (2*S*)-aziridine-2-carboxylate (**6**) shown in the following Scheme 1.

The aziridine carboxylate **6** was reacted with *N*,*O*-dimethylhydroxylamine in the presence of *i*-PrMgBr at 0 °C to form the Weinreb's amide **7** in 92% yield followed by addition of 1-pentadecynyl lithium to yield **8** in quantitative yield. Acyl aziridine **8** was selectively reduced to *erythro*-amino alcohol **9** to have the right stereochemistry of ceramide with $NaBH_4$ and $ZnCl_2$ via the chelation-controlled transition state in 98% yield.[11] Then alkyne **9** was reduced by LAH to yield *N*-[(*R*)-α-methylbenzyl]aziridin-2(*S*)-yl-hydroxy-1-pentadecene in 86% yield whose hydroxy group was protected by TBDMS in 93% yield. The aziridine ring was regioselectively opened by the reaction with acetic acid in CH_2Cl_2 to give acyclic compound **10** in 73%. The *O*-acetyl group in **10** was hydrolyzed by KOH in ethanol to yield the free alcohol at which stage oxazoldin-2-one ring was introduced by the reaction with 1,1'-carbodiimidazole in 70% yield for two steps. The α-methylbenzyl group attached on the nitrogen was removed by Na in liq. NH_3 at −78 °C to give the backbone ring **11** in 77% yield. The parent ring (**11**) was readily acylated on the nitrogen using acid chloride in the presence of a suitable base. Sodium hexamethyldisilazane was the most effective base being applicable to various acyl chlorides. The target molecules (**5a**–**5m**) were prepared from the parent ring compound **11** by the same reaction sequence with the corresponding acid chlorides followed by desilylations in over 85–95% yield in all cases over two steps.

All of these compounds were evaluated as potential antileukemic agents against human leukemia HL-60 cells with C2-Cer and C2-DHCer as positive and negative references, respectively by MTT assay whose results were shown in Figure 3.[22] Human leukemia HL-60 cells were treated with 20 μM of each constrained ceramide analogs with references for 16 h. All tested compounds (**5a**–**5m**) showed activities with drastic difference by the *N*-acyl

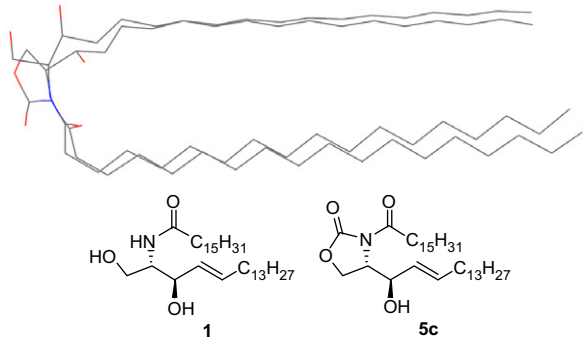
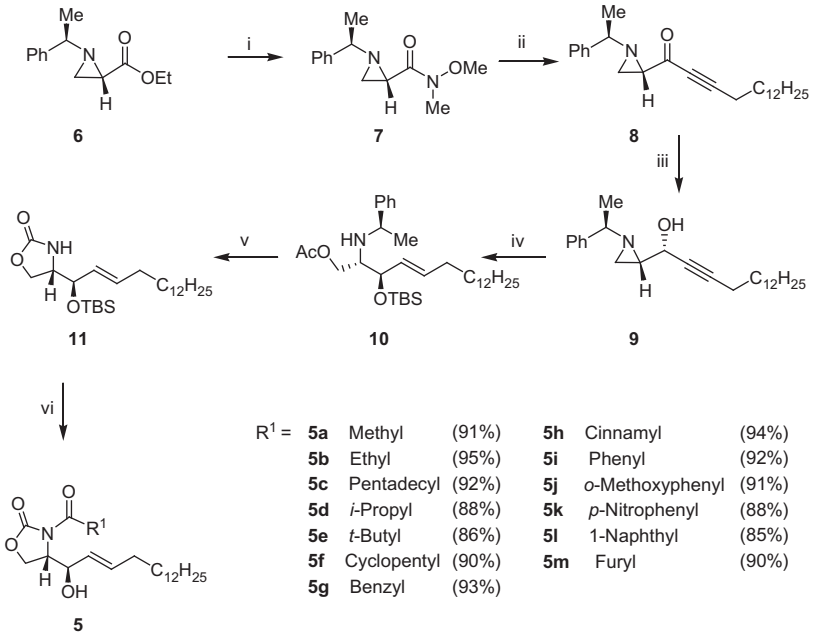


Figure 2. Overlay of naturally occurring C16-ceramide (**1**) and its oxazolidin-2-one analog (**5c**). Calculation was carried out with the commercial product Spartan® 04. The overlay structures were deduced by locking the bonds associated with nitrogen for comparison.



Scheme 1. Reagents and conditions: (i) $NH(OMe)CH_3$, *i*-PrMgBr, THF, 0 °C (92%); (ii) $HCCHCH_2C_{12}H_{25}$, *n*-BuLi, −78 °C then 0 °C, 2 h (99%); (iii) $ZnCl_2$, $NaBH_4$, MeOH, −78 °C, then −15 °C, 1 h (98%); (iv) (a) LAH, 0–60 °C, 8 h (86%); (b) DMAP, *t*-$BuMe_2SiCl$, rt (93%); (c) AcOH, CH_2Cl_2, rt, (73%); (v) (a) KOH, EtOH, rt, 1 h (81%); (b) CDI, DBU, rt, 24 h (87%); (c) Na, NH_3, −78 °C (77%); (vi) (a) R^1COCl, NaHMDS, 0 °C; (b) Bu_4NF, THF, 0 °C.

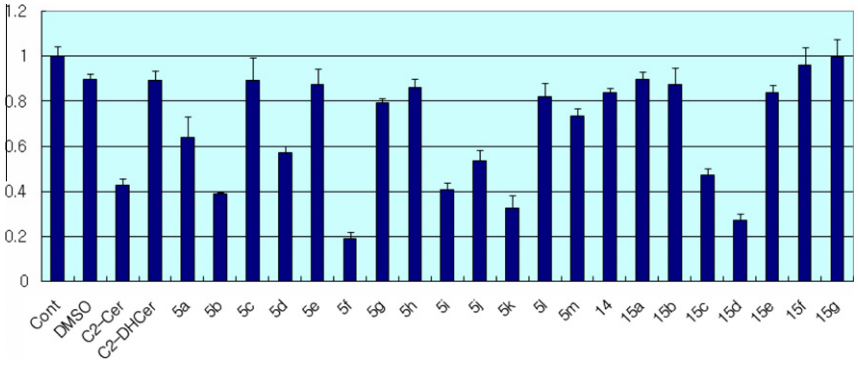


Figure 3. Relative cell survival of HL-60 cells after treatment with 20 μM of C2-Cer, C2-DHCer and oxazolidin-2-ones (**5a**–**5m**) and (**14**, **15a**–**15g**) for 16 h.

substituents of the oxazolin-2-one ring. *N*-acetyl compound **5a** mimicking C2-Cer has much lower activity than C2-Cer itself while a great deal of improvement was observed in the compound **5b** with additional one carbon as ethyl group in R^1 of the compound **5**. Compound **5c** with a long alkyl chain did not show any activity due to low cell permeability to produce its antileukemic activity that was also observed in natural C16-Cer.[16] The longer carbon chain than ethyl (**5b**) such as *i*-propyl (**5d**) or butyl (**5e**) did not improve the activity much. The cyclopropanyl ring at R^1 in the compound **5f** improved the activity drastically which showed the best activity among all tested compounds with alkyl substituents. Compounds with benzyl (**5g**) and cinnamyl (**5h**) were not active. Some of the compounds possessing benzoyl (**5i**–**5k**) rather than alkanoyl group (**5a**–**5h**) showed good activities. Their activities were dependent to the substituents of phenyl ring. Especially the compound (**5k**) with *p*-nitrophenyl for R^1 showed very potent activity. Aryl substituents other than phenyl for R^1 group in the compounds **5** did not improve the activity among the tested compounds including naphthyl (**5l**) and furyl (**5m**) substituents. The origin of the activity difference among all tested compounds is speculated not from the simply physical properties such as lipophilicity or polarity but from the unique structural features of each compound.

We decided to find out whether the cytotoxicity induced by these analogs or the reduction of HL60 cells was caused by the apoptosis. The characteristics of cell death were verified by the analysis of DNA-fragmentation induced by these molecules. DNA-laddering of fragments was observed after 15 h of incubation of HL-60 cells with 20 μM of C2-Cer, C2-DHCer and oxazolidin-2-ones (**5a**, **5b**, **5c**, **5d**, **5f**, **5i** and **5k**) shown in Figure 4. The picture showed the clear co-relationship between anticancer activity and DNA-fragmentation.

The active compounds such as **5b** (R^1 = ethyl) and **5i** (R^1 = phenyl) showed similar or a little more amount of DNA fragmentation as white bands than for C2-Cer while inactive **5a** (R^1 = methyl) and **5c** (R^1 = pentadecyl) had similar band to C2-DHCer's on the picture. All active compounds **5d** (R^1 = *i*-propyl), **5f** (R^1 = cyclopentyl), **5i** (R^1 = phenyl) and **5k** (*p*-nitrophenyl) also showed DNA-fragmented bands. This demonstrates cell death was caused by the apoptotic process. A good correlation between cell death of HL-60 cells and DNA fragmentation is solid evidence that the cytotoxicity of the constrained ceramide analogs is originated from apoptosis.

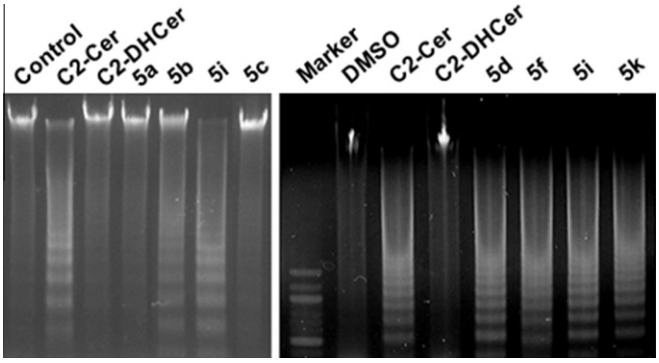


Figure 4. DNA fragmentation of HL-60 cells by the treatment with 20 μM of C2-Cer, C2-DHCer and some representative oxazolidin-2-ones (**5a**, **5b**, **5c**, **5d**, **5f**, **5i**, **5k**).

The cellular concentration of ceramide, measured three times by the reported method[23] after the treatment of the active analogs, showed minor changes by less than 15%, which means the oxazolidin-2-one ceramide analogs did not inhibit the ceramide metabolic enzymes to accumulate the cellular concentration of ceramide. Instead these molecules induced apoptosis through caspase cascade with induction of nuclear factor κB (NF-κB) DNA binding and caspase-3 activation as exogeneous C-2 and C-6-ceramides do.[16b]

All of the compounds synthesized and tested up to this point have 1-hydroxyhexadec-2-ene at C4 of oxazolidin-2-one[24] which is the same backbone with eighteen carbons as the sphingoid's. For the investigation of the effect by changing the sphingoid backbone were designed and synthesized many analogs bearing different carbon numbers with and without ring. They have different sphingoid backbones consisted of different carbon chains at R^2 in Scheme 2 bearing the olefin at the right position that the natural sphingoid has. The cyclopentanoyl group was attached at the ring nitrogen because it showed the best biological activity among other substituent. Those included compounds having four (**15a**), six (**15b**), eight (**15c**) and ten (**15d**) straight carbons for the R^2 group. Others are cyclopentyl (**15e**), cyclohexyl (**15f**) and benzyl (**15g**) groups. All of these were synthesized from the methathesis[25] between the common intermediate **14** and the corresponding olefins including 1-hexene, 1-octene, 1-decene, 1-dodecene, vinylcyclopentane, vinylcyclohexane and allylbenzene in the presence of the Grubbs's 2nd generation catalyst in high yields (Scheme 2).

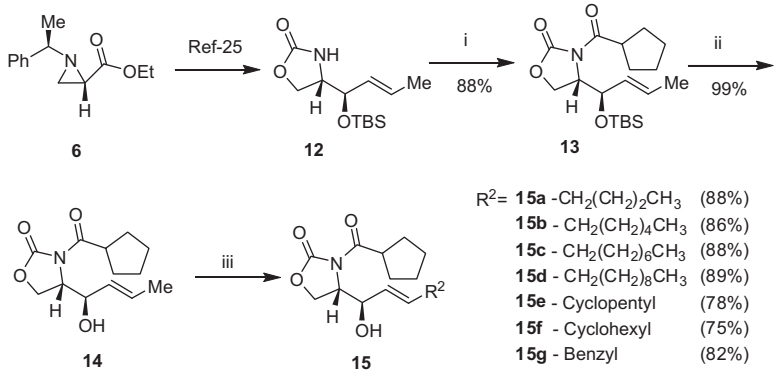
All of them except **15d** showed the worse activities compared to the C2-Cer. There is a drastic change of the activities by the carbon chain length corresponding to the sphingoid backbone. Almost no activity was observed from the compounds (**15a**–**15c**) bearing the chain length less than 12 in R^2. Only **15d** consisted of 15 carbons at the backbone, which is shorter than natural ceramide by three carbon units, shows comparably good activity. The compounds with cyclopentyl, cyclohexyl and benzyl at the backbone site showed no activity either. This indicates that the hydrophobic binding site of the receptor inducing anticancer activity is quite deep enough to accommodate carbon chains with certain length and is narrow enough not to get the cyclic compounds. This is the first detailed study on the structure and activity relationship in the pro-apoptotic process with diverse oxazolidin-2-ones as constrained ceramide analogs.

3. Conclusion

Heterocyclic analogs of ceramide as 3-alkanoyl or benzoyl-4-(1-hydroxy-2-enyl)-oxazolidin-2-ones were designed by binding of primary alcohol and amide in sphingosine backbone as a carbamate. These were based on the unique butterfly conformer of natural ceramide folded by the strong internal hydrogen bonding between amide and two alcohols at C1 and C3 and by the hydrophobic interaction between two long alkyl chain of sphingosine backbone and fatty acid. Some of them with different acyl group at the ring nitrogen were synthesized by addition of acyl halide to the common ring 4-(1-*t*-butyldimethylsilyloxyhexadec-2-enyl)-oxazolidin-2-one which was elaborated from chiral aziridine-2-carboxylate including stereoselective reduction and ring opening reactions. Other analogs with different carbon backbone at C4 were prepared from 3-cyclopentanecarbonyl-4-(1-*t*-butyldimethylsilyloxybut-2-enyl)-oxazolidin-2-one by cross metathesis. All compounds were tested as antileukemic drugs against human leukemia HL-60 cells. Many of them including propionyl, cyclopentanoyl and *p*-nitrobenzoyl-4-(1-hydroxyhexadec-2-enyl)-oxazolidin-2-ones showed better antileukemic activities than natural C2-Cer with good correlation between cell death and DNA fragmentation. There is a drastic change of the activities by the carbon chain lengths at C4 position which is corresponding to the sphingoid backbone. Almost no activity was observed from the compounds bearing the chain length less than twelve of sphingoid backbone. Cytotoxicity was induced by caspase activation without significant accumulation of endogenous ceramide concentration or any perturbation of ceramide metabolism.

4. Experimental

All non-aqueous reactions were run in flame-dried glassware under a positive pressure of nitrogen with exclusion of moisture from reagents and glassware using standard techniques for



Scheme 2. Reagents and conditions: (i) Sodium bis(trimethylsilyl)amide, cyclopentanecarbonyl chloride, THF, −78 °C, 2 h; (ii) TBAF, THF, 0 °C, 1 h, then at rt, 2 h; (iii) benzylidene[1,3-bis(2,4,6-trimethylphenyl)-2-imidazolidinylidene]dichloro(tricyclohexylphosphine)ruthenium, CH_2CH_2, 24 h.

manipulating air-sensitive compounds. Anhydrous solvents were obtained using standard drying techniques. Unless stated otherwise and commercial grade reagents were used without further purification. (2*S*)-1-[(*R*)-α-methylbenzyl]-2-aziridinecarboxylate ethyl esters were purchased from Imagene. Their menthol esters were also purchased from Aldrich. Reactions were monitored by analytical thin-layer chromatography (TLC) performed on precoated, glass-backed silica gel plates. Visualization of the developed chromatogram was performed by UV absorbance, ninhydrin, phosphomolybdic acid or iodine. Flash chromatography was performed on 230–400 mesh silica gel with the indicated solvent systems. Melting points are uncorrected. Routine nuclear magnetic resonance spectra were recorded either on Varian Gemini 200 (200 MHz) or Varian Gemini 400 (400 MHz) spectrometers. Chemical shifts for ^{1}H NMR spectra are recorded in parts per million from tetramethylsilane with the solvent resonance as the internal standard ($CDCl_3$, δ 7.27 ppm and CD_3OD δ 3.31 ppm). Data are reported as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, qn = quintet, m = multiplet, and br = broad), coupling constant in Hertz, and integration. Chemical shifts for ^{13}C NMR spectra are recorded in parts per million from tetramethylsilane using the central peak of the solvent resonance as the internal standard ($CDCl_3$, δ 77.00 ppm and CD_3OD δ 49.15). All spectra were obtained with complete proton decoupling.

4.1. 1-{(*S*)-1-[(*S*)-1-phenylethyl]aziridin-2-yl}hexadec-2-yn-1-one (8)[11]

To a solution of pentadecyne (1.5 equiv) in anhydrous THF (0.3 M) under nitrogen at −78 °C was added *n*-BuLi (1.3 equiv). The mixture was stirred for 30 min, and then treated with **7** (1 g, 4.27 mmol) via canula at −78 °C. The mixture was stirred for 1 h at −78 °C and was warmed to room temperature and then treated with 10 mL of water. The organic layer was separated and the aqueous layer was extracted with EtOAc (20 mL × 4). The combined organic extracts were dried over anhydrous $MgSO_4$, filtered, and concentrated in vacuo. Purification by silica gel flash chromatography gave 1.45 g of product **8** in 99% yield.

4.2. (*R*)-1-{(*S*)-1-[(*R*)-1-phenylethyl]aziridin-2-yl}hexadec-2-yn-1-ol (9)[11]

To the solution of **8** (1 g, 2.62 mmol) in MeOH (0.3 M) at −78 °C was added $ZnCl_2$ (1.2 equiv). The solution was stirred for 30 min, and $NaBH_4$ (2 equiv) was added at −78 °C. The mixture was stirred for 30 min, and 20 mL of water was added. The organic layer was separated. The aqueous layer was extracted with CH_2Cl_2, and the combined organic extracted were dried, filtered, and concentrated in vacuo. Purification by silica gel flash chromatography provided **9** in 98% yield.

4.3. (*E*,2*S*,3*R*)-2-[(*R*)-1-Phenylethylamino]-3-(*t*-butyldimethylsilyloxy)-octadec-4-enyl acetate (10)

To a stirred solution of substrate in anhydrous THF was added LAH at room temperature. After 3 h, the reaction was completed according to TLC, quenched by aqueous $KHSO_4$, extracted with EtOAc/H_2O, dried over $MgSO_4$, and purified by column chromatography to give the reaction product in 86% yield. The stirred solution of the reduced substrate in CH_2Cl_2 was added DMAP at room temperature. The mixture was added TBDMSCl at room temperature. The reaction mixture was stirred for 17 h at room temperature and was treated with 10 mL of saturated $NaHCO_3$ solution. The organic extracts were washed with brine, dried over anhydrous $MgSO_4$, filtered, and concentrated in vacuo. Purification by silica flash chromatography gave product in 93% yield.[11] To a solution of substrate obtained above reaction in CH_2Cl_2 under a nitrogen atmosphere at room temperature was added AcOH. The mixture was stirred for 10 h and then quenched with 10 mL of saturated $NaHCO_3$ solution. The organic layer was separated and the aqueous layer was extracted with CH_2Cl_2. The combined organic extracts were washed with brine, dried over anhydrous $MgSO_4$, filtered, and concentrated in vacuo. Purification by column chromatography gave **10** in 73% yield: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.29–7.22 (m, 5H), 5.62 (dt, *J* = 15.4 Hz, 1H), 5.30 (dd, *J* = 14.7 Hz, 1H), 4.16–4.11 (m, 3H), 3.91 (q, *J* = 6.6 Hz, 1H), 2.52–2.49 (m, 1H), 2.10–1.98 (m, 5H), 1.29–1.19 (m, 25H), 0.85 (s, 12H), 0.00 (s, 3H), −0.02 (s, 3H); HRMS (ESI) calcd for $[C_{34}H_{61}NO_3Si+H]^+$: 560.4499, found 560.4504.

4.4. (*S*)-4-[(*R*,*E*)-1-*t*-Butyldimethylsilyloxyhexadec-2-enyl]oxazolidin-2-one (11)

To a solution of **10** in EtOH at room temperature was added KOH. The mixture was stirred for 1 h at room temperature and then quenched with 10 mL of water. The product was extracted with CH_2Cl_2 and the combined organic extracts were washed with brine, dried over $MgSO_4$, filtered, and concentrated. Purification by column chromatography gave product in 81% yield. Then this was dissolved in CH_2Cl_2. The resultant solution was stirred followed by adding DBU and carbonyl diimidazole at room temperature. The mixture was stirred for overnight and then concentrated in vacuo. Purification by silica gel flash chromatography gave product in 87% yield. Anhydrous NH_3 was condensed into a flask containing a lithium metal maintained at −78 °C until a dark blue color persisted and then added *tert*-butyl alcohol. The mixture was stirred for 5 min at −78 °C and then added substrate in THF. The resulting mixture was stirred for 30 min and then quenched with water. The mixture was extracted with EtOAc. The combined extracts were washed with brine, dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Purification by silica gel flash chromatography gives product **11** in 77% yield: ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.64 (dt, *J* = 15, 5 Hz, 1H), 5.59 (s, 1H), 5.38 (dd *J* = 15, 5 Hz, 1H), 4.32 (dd, *J* = 19, 9 Hz, 1H), 4.29–4.27 (m, 1H), 4.00 (t, *J* = 6 Hz, 1H), 3.89–3.84 (m, 1H), 2.06–2.01 (m, 2H), 1.41–1.38 (m, 22H), 0.92 (s, 12H), 0.07 (s, 3H), 0.04 (s, 3H); HRMS (ESI) calcd for $[C_{25}H_{49}NO_3Si+H]^+$: 440.3560, found 440.3569.

4.5. General procedure for acylation and desilylation procedure (5)

To a stirred solution of **11** in anhydrous THF was added sodium bis(trimethylsilyl)amide (1.2 equiv) at −78 °C. After 10 min at −78 °C, the mixture was added acylchloride (1.3 equiv) at −78 °C. The reaction was complete according to TLC, extracted with EtOAc/H_2O, dried over $MgSO_4$, and purified by column chromatography to give the coupled product. A solution of substrate in anhydrous THF was added TBAF at 0 °C. After 4 h, the reaction mixture was complete, extracted with EtOAc/H_2O, dried over $MgSO_4$, and purified by column chromatography to give final product (**5**) in 85–95% yield after two steps.

4.5.1. (*S*)-3-Acetyl-4-[(*R*,*E*)-1-hydroxyhexadec-2-enyl]oxazolidin-2-one (5a)

Yield: 91%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.86 (dt, *J* = 11.2, 4 Hz, 1H), 5.39–5.25 (m, 2H), 4.45 (t, *J* = 9 Hz, 1H), 4.30–4.28 (m, 1H), 4.04–4.01 (m, 1H), 2.06 (q, *J* = 5 Hz, 2H), 1.48–1.39 (m, 23H), 1.12 (t, *J* = 7 Hz, 3H), 0.88 (t, *J* = 5 Hz, 3H); HRMS (ESI) calcd for $[C_{21}H_{37}NO_4+H]^+$: 368.2801, found 368.2814.

4.5.2. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-propionyloxazolidin-2-one (5b)

Yield: 95%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.88 (dt, J = 11, 4 Hz, 1H), 5.23–5.32 (m, 2H), 4.47 (t, J = 9 Hz, 1H), 4.31–4.29 (m, 1H), 4.04–4.01 (m, 1H), 2.35 (q, J = 5 Hz, 2H), 2.08 (q, J = 5 Hz, 2H), 1.41–1.36 (m, 23H), 1.14 (t, J = 7 Hz, 3H), 0.89 (t, J = 5 Hz, 3H); HRMS (ESI) calcd for $[C_{22}H_{39}NO_4+H]^+$: 382.2957, found 382.2951.

4.5.3. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-palmityryloxazolidin-2-one (5c)

Yield: 92%; ^{1}H NMR (300 MHz, $CDCl_3$) 5.88 (dt, J = 12, 4 Hz, 1H), 5.19–5.33 (m, 3H), 4.44 (t, J = 9 Hz, 1H), 4.26 (m, 1H), 4.01 (m, 1H), 2.34 (t, J = 7 Hz, 2H), 2.07 (q, J = 7 Hz, 2H), 1.70–1.68 (m, 3H), 1.28–1.18 (m, 48H), 0.89 (t, J = 7 Hz, 3H); HRMS (ESI) calcd for $[C_{35}H_{65}NO_4+H]^+$: 564.4992, found 564.4983.

4.5.4. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(isobutyryl)oxazolidin-2-one (5d)

Yield: 88%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.88 (dt, J = 8.4, 6.6 Hz, 1H), 5.4–5.25 (m, *2*H), 4.43 (t, J = 9.0 Hz, 1H), 4.26 (dd, J = 9.0, 5.1 Hz, 1H), 4.03–3.97 (m, 1H), 2.63–2.53 (m, 1H), 2.08 (d, J = 6.0 Hz, 1H), 2.03 (d, J = 6.6 Hz, 2H), 1.4–1.2 (m, 22H), 1.19 (d, J = 3.3 Hz, 3H), 1.17 (d, J = 3.3 Hz, 3H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 176.0, 159.1, 138.7, 122.2, 73.9, 65.9, 54.6, 34.1, 32.3, 31.9, 29.6, 29.5, 29.4, 29.3, 29.2, 29.1, 28.7, 22.6, 18.9, 18.7, 14.0; HRMS (ESI) calcd for $[C_{23}H_{41}NO_4+H]^+$: 396.3114, found 396.3121.

4.5.5. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(pivaloyl)oxazolidin-2-one (5e)

Yield: 86%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.91–5.81 (m, 1H), 5.44 (br s, 1H), 5.37–5.27 (m, 2H), 4.42 (t, J = 9.0 Hz, 1H), 4.27 (dd, J = 9.0, 5.1 Hz, 1H), 4.03–4.96 (m, 1H), 2.08 (d, J = 6.6 Hz, 1H), 2.03 (d, J = 4.5 Hz, 2H), 1.36–1.22 (m, 22H), 1.21 (s, 9H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 169.8, 138.4, 122.2, 73.7, 70.5, 65.8, 54.7, 32.3, 31.9, 29.6, 29.4, 29.0, 28.7, 27.0, 22.6, 14.1; HRMS (ESI) calcd for $[C_{24}H_{43}NO_4+H]^+$: 410.3270, found 410.3268.

4.5.6. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(cyclopentyl)oxazolidin-2-one (5f)

Yield: 90%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 5.9–5.8 (m, 1H), 5.44 (br s, 1H), 5.4–5.2 (m, 2H), 4.45 (t, J = 9.0 Hz, 1H), 4.28 (dd, J = 9.0, 5.1 Hz, 1H), 4.1–3.9 (m, 1H), 2.8–2.7 (m, 1H), 2.11 (d, J = 6.3 Hz, 1H), 2.06 (d, J = 7.5 Hz, 1H), 2.0–1.6 (m, 8H), 1.28 (br s, 22H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 175.7, 159.0, 138.7, 122.3, 73.9, 65.9, 54.7, 43.9, 32.3, 31.9, 29.6, 29.5, 29.4, 29.3, 29.2, 29.1, 29.7, 25.8, 25.7, 22.6, 14.1; HRMS (ESI) calcd for $[C_{25}H_{43}NO_4+H]^+$: 422.3270, found 422.3277.

4.5.7. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(2-phenylacetyl)oxazolidin-2-one (5g)

Yield: 93%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.4–7.2 (m, 5H), 5.86–5.76 (m, 1H), 5.34–5.22 (m, 2H), 5.09 (br s, 1H), 4.35 (t, J = 9.0 Hz, 1H), 4.12 (dd, J = 9.0, 5.1 Hz, 1H), 3.98–3.92 (m, 1H), 3.65 (s, *2*H), 2.02 (q, J = 6.9 Hz, 2H), 1.35–1.24 (m, 22H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 169.9, 139.4, 133.5, 129.1, 128.7, 127.3, 121.7, 74.8, 65.9, 54.4, 41.4, 39.7, 32.3, 31.9, 29.6, 29.5, 29.4, 29.3, 29.2, 29.1, 28.7, 22.6, 14.1; HRMS (ESI) calcd for $[C_{27}H_{41}NO_4+H]^+$: 444.3114, found 444.3121.

4.5.8. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(cinnamyl)oxazolidin-2-one (5h)

Yield: 94%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.71 (d, J = 16.2 Hz, 1H), 7.55–7.5 (m, 2H), 7.45–7.38 (m, 3H), 6.43 (d, J = 16.2 Hz, 1H), 6.0–5.9 (m, 1H), 5.5–5.3 (m, 2H), 5.27 (br s, 1H), 4.48 (t, J = 9.0 Hz, 1H), 4.30 (dd, J = 9.0, 5.1 Hz, 1H), 4.12–4.0 (m, 1H), 2.10 (d, J = 5.1 Hz, 1H), 2.05 (d, J = 7.5 Hz, 1H), 1.25 (br s, 22H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.1, 146.4, 139.8, 129.2, 128.5, 122.3, 117.4, 75.0, 66.5, 54.9, 32.7, 32.2, 30.0, 29.9, 29.6, 29.4, 29.0, 22.9, 14.4; HRMS (ESI) calcd for $[C_{28}H_{41}NO_4+H]^+$: 456.3114, found 456.3118.

4.5.9. (*S*)-3-Phenyl-4-[(*R*,*E*)-1-hydroxyhexadec-2-enyl]oxazolidin-2-one (5i)

Yield: 92%; mp = 82 °C; ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.04 (d, J = 8 Hz, 2H), 7.59 (t, J = 8 Hz, 1H), 7.46 (t, J = 8 Hz, 2H), 5.96 (m, 2H), 5.55 (m, 2H), 4.49 (t, J = 8 Hz, 1H), 4.38 (m, 1H), 4.05 (m, 1H), 2.08 (q, J = 7 Hz, 2H), 1.24 (m, 22H), 0.89 (t, J = 7 Hz, 3H); HRMS (ESI) calcd for $[C_{26}H_{39}NO_4+H]^+$: 430.2957, found 430.2954.

4.5.10. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(2-tolyl)oxazolidin-2-one (5j)

Yield: 91%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.88 (dd, J = 4.8, 1.8 Hz, 1H), 7.52 (td, J = 9.0, 1.8 Hz, 1H), 7.1–6.9 (m, 2H), 6.1–5.9 (m, 2H), 5.6–5.4 (m, 3H), 4.51 (t, J = 9.0 Hz, 1H), 4.37 (dd, J = 9.0, 4.8 Hz, 1H), 4.2–4.1 (m, 1H), 3.94 (s, 3H), 2.12 (d, J = 6.6 Hz, 1H), 2.07 (d, J = 7.2 Hz, 1H), 1.4–1.2 (m, 21H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 165.4, 159.2, 158.9, 151.3, 139.3, 133.9, 131.7, 121.9, 120.3, 112.0, 74.8, 66.1, 55.9, 54.6, 32.4, 31.9, 29.6, 29.5, 29.4, 29.3, 29.2, 29.1, 29.7, 22.7, 14.1; HRMS (ESI) calcd for $[C_{27}H_{41}NO_5+H]^+$: 460.3063, found 460.3070.

4.5.11. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(4-nitrophenyl)-oxazolidin-2-one (5k)

Yield: 88%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.28 (dd, J = 6.9, 1.8 Hz, 2H), 8.20 (dd, J = 6.9, 1.8 Hz, 2H), 6.1–5.9 (m, *2*H), 5.6–5.4 (m, 2H), 4.53 (t, J = 9.0 Hz, 1H), 4.37 (dd, J = 9.0, 4.5 Hz, 1H), 4.2–4.1 (m, 1H), 2.09 (q, J = 6.9 Hz, 2H), 1.4–1.2 (m, *22*H), 0.88 (t, J = 6.9 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 163.6, 159.2, 140.4, 134.8, 130.8, 123.7, 121.3, 99.9, 76.4, 65.9, 54.6, 32.4, 31.9, 29.6, 29.5, 29.4, 29.3, 29.2, 29.1, 28.6, 22.6, 14.1; HRMS (ESI) calcd for $[C_{26}H_{38}N_2O_6+H]^+$: 475.2808, found 475.2815.

4.5.12. (*S*)-4-[(*R*,*E*)-1-Hydroxyhexadec-2-enyl]-3-(1-naphthoyl)-oxazolidin-2-one (5l)

Yield: 85%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.89 (d, J = 7.2 Hz, 1H), 8.20 (d, J = 7.2 Hz, 1H), 8.07 (d, J = 7.1 Hz, 1H), 7.92 (d, J = 7.1 Hz, 1H), 7.68–7.51 (m, 4H), 6.06 (dt, J = 15.0, 6.6 Hz, 1H), 5.76 (br s, 1H), 5.64–5.50 (m, 2H), 4.55 (t, J = 9.0 Hz, 1H), 4.40 (dd, J = 9.0, 4.8 Hz, 1H), 4.25–4.19 (m, 1H), 2.12 (dd, J = 13.8, 6.9 Hz, 2H), 1.45–1.25 (m, 21H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 172.5, 166.5, 159.4, 139.9, 134.1, 131.7, 130.6, 128.9, 128.3, 126.6, 125.8, 124.8, 122.4, 75.5, 66.5, 55.1, 32.7, 32.2, 29.9, 29.8, 29.7, 29.6, 29.5, 29.4, 29.0, 22.9, 14.4; HRMS (ESI) calcd for $[C_{30}H_{41}NO_4+H]^+$: 480.3114, found 480.3120.

4.5.13. (*S*)-3-(Furan-2-carbonyl)-4-((*R*,*E*)-1-hydroxyhexadec-2-enyl)oxazolidin-2-one (5m)

Yield: 90%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.62 (d, J = 0.6 Hz, 1H), 7.27 (d, J = 3.3 Hz, 1H), 6.55 (dd, J = 3.3, 1.5 Hz, 1H), 6.06–5.95 (m, 1H), 5.86 (br s, 1H), 5.52–5.40 (m, 2H), 4.51 (t, J = 3.3 Hz, 2H), 4.33 (dd, J = 9.0, 4.2 Hz, 1H), 4.2–4.1 (m, 2H), 2.12 (d, J = 6.9 Hz, 1H), 2.08 (d, J = 6.9 Hz, 1H), 1.45–12.5 (m, 20H), 0.88 (t, J = 6.6 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 169.9, 159.1, 157.5, 146.8, 143.9, 140.0, 121.8, 118.8, 112.0, 75.3, 66.1, 54.6, 32.4, 31.9, 29.6, 29.5, 29.4, 29.3, 29.1, 28.7, 22.7, 14.1; HRMS (ESI) calcd for $[C_{24}H_{37}NO_5+H]^+$: 420.2750, found 420.2752.

4.5.14. (*S*)-4-[(*R*,*E*)-1-(*t*-Butyldimethylsilyloxy)but-2-enyl]-3-cyclopentanecarbonyl)oxazolidin-2-one (13)

To a stirred solution of **12** (2.0 g, 7.37 mmol) in anhydrous THF (22.1 mL) at −78 °C, under an inert atmosphere of N_2, were added

sodium bis(trimethylsilyl)amide (1 M in THF solution, 44.2 mL). The solution was stirred for 30 min, and cyclopentanecarbonyl chloride (3.58 mL, 27.0 mmol) was added at −78 °C. After stirring at −78 °C for 2 h. The reaction mixture was warmed to room temperature and quenched with aqueous H_2O (20 mL). The reaction product was extracted with ethyl acetate (3 × 50 mL) and the combined organic layers were washed with brine, dried ($MgSO_4$), filtered and concentrated in vacuo. The residue was purified by flash column chromatography on silica gel (*n*-hexane/EtOAc = 90:10) to give **13** (2.38 g, 88%) as white oil: ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.81 (dq, J = 14.3, 6.5 Hz, 1H), 5.33 (ddq, J = 15.3, 7.2, 1.6 Hz, 1H), 4.65 (dd, J = 5.5, 1.3 Hz, 1H), 4.45 (dd, J = 8.0, 3.0 Hz, 1H), 4.36 (ddq, J = 8.4, 2.8, 1.7 Hz, 1H), 4.16 (t, J = 8.2 Hz, 1H), 4.0–3.81 (m, 1H), 2.05–1.6 (m, 11H), 0.88 (s, 9H), −0.026 (s, 3H), −0.044 (s, 3H); ^{13}C NMR (50 MHz; $CDCl_3$) δ −5.7, −4.9, 17.2, 17.3, 25.1, 25.5, 25.5, 29.2, 29.9, 42.4, 58.1, 61.4, 66.8, 128.0, 128.7, 153.1, 176.2; HRMS (ESI) calcd for $[C_{19}H_{33}NO_4Si+Na]^+$: 390.2077, found 390.2076.

4.5.15. (*S*)-3-(Cyclopentanecarbonyl)-4-[(*R*,*E*)-1-hydroxybut-2-enyl]oxazolidin-2-one (14)

To a solution of **13** (1.0 g, 2.72 mmol) in anhydrous THF (8.16 mL) at 0 °C, under an inert atmosphere of N_2, was added TBAF (2.72 mL, 2.72 mmol, 1 M in THF). The reaction mixture was then stirred for 1 h at 0 °C and warmed to room temperature and stirred for an additional 2 h before being quenched with saturated aqueous NH_4Cl solution (5.0 mL). The reaction mixture was extracted with ethyl acetate (3 × 20 mL) and combined organic layers were washed with brine, dried ($MgSO_4$), filtered and concentrated in vacuo. The residue was purified by flash column chromatography on silica gel (*n*-hexane/EtOAc = 75:25) to give **14** (0.68 g, 99%) as white solid: mp = 42 °C; ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.88 (dq, J = 15.7, 7.6 Hz, 1H), 5.84 (br s, 1H), 5.36 (ddq, J = 14.8, 7.3, 1.6 Hz, 1H), 5.25 (dd, J = 7.3, 4.2 Hz, 1H), 4.41 (t, J = 8.9 Hz, 1H), 4.23 (dd, J = 8.9, 5.0 Hz, 1H), 3.98 (ddd, J = 13.9, 8.9, 4.5 Hz, 1H), 2.84–2.65 (m, 1H), 1.97–1.52 (m, 11H); ^{13}C NMR (50 MHz; $CDCl_3$) δ 17.9, 25.7, 25.7, 29.7, 29.9, 43.8, 54.7, 65.9, 74.0, 124.0, 132.6, 159.9, 175.8; HRMS (ESI) calcd for $[C_{13}H_{19}NO_4+Na]^+$: 276.1212, found 276.1211.

4.6. General procedure for the preparation of the compound 15

To a solution of **14** (1 equiv) in anhydrous dichloromethane (0.48 M) at room temperature under an inert atmosphere of N_2 was added the corresponding olefin (1.2 equiv). Specifically for the preparation of **15a**, **15b**, **15c**, **15d**, **15e**, **15f** and **15g** 1-hexene, 1-octene, 1-nonene, 1-dodecene, methylenecyclopentane, methylenecyclohexane and stylene were used. The reaction mixture was then stirred for 5 min. Grubbs type II catalyst (benzylidene[1,3-bis(2,4,6-trimethylphenyl)-2-imidazolidinylidene]dichloro(tricyclohexylphosphine)-ruthenium, 5 mol %) was added in one portion, and resulting homogeneous solution was stirred for 24 h at room temperature. After evaporation, the residue was purified by flash column chromatography on silica gel (*n*-hexane/EtOAc = 75:25, v/v) to give **15** in 75–89% yield.

4.6.1. (*S*)-3-(Cyclopentanecarbonyl)-4-[(*R*,*E*)-1-hydroxyhept-2-enyl]oxazolidin-2-one (15a)

Yield: 88%; ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.89 (ddd, J = 20.7, 13.6, 6.5 Hz, 1H), 5.63 (br s, 1H), 5.41–5.23 (m, 2H), 4.43 (t, J = 8.8 Hz, 1H), 4.25 (dd, J = 8.8, 5.0 Hz, 1H), 4.00 (dd, J = 8.7, 4.3 Hz, 1H), 2.87–2.65 (m, 1H), 2.08 (q, J = 6.7 Hz, 2H), 1.94–1.56 (m, 8H), 1.40–1.24 (m, 4H), 0.89 (t, J = 6.8 Hz, 3H); ^{13}C NMR (50 MHz; $CDCl_3$) δ 13.5, 22.8, 25.4, 25.5, 29.5, 29.7, 30.6, 31.7, 43.6, 54.5, 65.6, 73.7, 122.3, 137.7, 159.6, 175.4; HRMS (ESI) calcd for $[C_{16}H_{25}NO_4+Na]^+$: 318.1681, found 318.1683.

4.6.2. (*S*)-3-(Cyclopentanecarbonyl)-4-[(*R*,*E*)-1-hydroxynon-2-enyl]oxazolidin-2-one (15b)

Yield: 86%; ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.87 (dq, J = 15.7, 7.6 Hz, 1H), 5.42–5.20 (m, 3H), 4.43 (t, J = 8.9 Hz, 1H), 4.25 (dd, J = 8.9, 5.0 Hz, 1H), 4.00 (dd, J = 8.7, 4.5 Hz, 1H), 2.89–2.57 (m, 1H), 2.05 (q, J = 6.9 Hz, 2H), 1.91–1.61 (m, 8H), 1.26 (br s, 8H), 0.88 (t, J = 6.8 Hz, 3H); ^{13}C NMR (50 MHz; $CDCl_3$) δ 13.9, 22.4, 25.5, 25.6, 28.5, 28.6, 29.7, 29.9, 31.4, 32.2, 43.7, 54.6, 66.8, 73.8, 122.3, 138.1, 159.5, 175.5; HRMS (ESI) calcd for $[C_{18}H_{29}NO_4+Na]^+$: 346.1994, found 346.1966.

4.6.3. (*S*)-3-(Cyclopentanecarbonyl)-4-[(*R*,*E*)-1-hydroxyundec-2-enyl]oxazolidin-2-one (15c)

Yield: 88%; ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.88 (dq, J = 15.7, 7.6 Hz, 1H), 5.65–5.15 (m, 3H), 4.43 (t, J = 8.9 Hz, 1H), 4.25 (dd, J = 8.9, 5.0 Hz, 1H), 4.00 (dd, J = 8.6, 4.3 Hz, 1H), 2.94–2.50 (m, 1H), 2.04 (q, J = 6.9 Hz, 2H), 1.95–1.45 (m, 8H), 1.26 (br s, 12H), 0.88 (t, J = 6.4 Hz, 3H); ^{13}C NMR (50 MHz; $CDCl_3$) δ 13.9, 22.5, 25.5, 25.6, 28.6, 18.9, 29.1, 29.2, 29.7, 29.8, 31.7, 32.2, 43.7, 54.6, 65.7, 73.8, 122.3, 138.1, 159.6, 175.5; HRMS (ESI) calcd for $[C_{20}H_{33}NO_4+Na]^+$: 374.2307, found 374.2305.

4.6.4. (*S*)-3-(Cyclopentanecarbonyl)-4-[(*R*,*E*)-1-hydroxytridec-2-enyl]oxazolidin-2-one (15d)

Yield: 89%; ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.87 (dq, J = 15.7, 7.6 Hz, 1H), 5.55–5.20 (m, 3H), 4.43 (t, J = 8.9 Hz, 1H), 4.25 (dd, J = 8.9, 5.0 Hz, 1H), 4.00 (dd, J = 8.6, 4.3 Hz, 1H), 2.87–2.64 (m, 1H), 2.05 (q, J = 6.9 Hz, 2H), 1.92–1.56 (m, 8H), 1.26 (br s, 16H), 0.88 (t, J = 6.4 Hz, 3H); ^{13}C NMR (50 MHz; $CDCl_3$) δ 13.9, 22.5, 25.5, 25.6, 28.6, 28.75, 28.9, 29.2, 29.2, 29.4, 29.6, 29.9, 31.7, 32.2, 43.7, 54.6, 65.7, 73.8, 122.3, 138.0, 159.6, 175.5; HRMS (ESI) calcd for $[C_{22}H_{37}NO_4+Na]^+$: 402.2620, found 402.2623.

4.6.5. (*S*)-3-(Cyclopentanecarbonyl)-4-[(*R*,*E*)-3-cyclopentyl-1-hydroxyallyl]oxazolidin-2-one (15e)

Yield: 78%; mp = 49 °C; ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.85 (dd, J = 14.3, 7.6 Hz, 1H), 5.67 (br s, 1H), 5.49–5.05 (m, 2H), 4.42 (t, J = 8.9 Hz, 1H), 4.24 (dd, J = 8.9, 5.0 Hz, 1H), 4.00 (dd, J = 9.0, 4.1 Hz, 1H), 2.90–2.59 (m, 1H), 2.58–2.17 (m, 1H), 1.89–1.51 (m, 12H), 1.25 (br s, 4H); ^{13}C NMR (50 MHz; $CDCl_3$) δ 24.9, 25.6, 25.7, 29.73, 29.9, 32.6, 32.7, 42.9, 43.8, 54.7, 65.9, 73.9, 120.3, 142.8, 159.4, 175.6; HRMS (ESI) calcd for $[C_{17}H_{25}NO_4+Na]^+$: 330.1681, found 329.9668.

4.6.6. (*S*)-4-[(*R*,*E*)-3-Cyclohexyl-1-hydroxyallyl]-3-(cyclopentanecarbonyl)oxazolidin-2-one (15f)

Yield: 75%; mp = 41 °C; ^{1}H NMR (200 MHz; $CDCl_3$) δ 5.82 (dq, J = 15.7, 7.6 Hz, 1H), 5.70 (br s, 1H), 5.42–5.09 (m, 2H), 4.41 (t, J = 8.9 Hz, 1H), 4.24 (dd, J = 8.9, 4.9 Hz, 1H), 4.00 (ddd, J = 12.9, 8.6, 4.3 Hz, 1H), 2.87–2.62 (m, 1H), 1.97–1.54 (m, 13H), 1.32–0.98 (m, 6H); ^{13}C NMR (50 MHz; $CDCl_3$) δ 25.5, 25.6, 25.8, 29.6, 29.8, 32.2, 40.2, 43.7, 54.6, 65.7, 73.9, 119.8, 143.3, 159.5, 175.5; HRMS (ESI) calcd for $[C_{18}H_{27}NO_4+Na]^+$: 344.1838, found 344.1836.

4.6.7. (*S*)-3-(Cyclopentanecarbonyl)-4-[(*R*,*E*)-1-hydroxy-4-phenylbut-2-enyl]oxazolidin-2-one (15g)

Yield: 82%; mp = 48 °C; ^{1}H NMR (400 MHz; $CDCl_3$) δ 7.65–6.76 (m, 5H), 5.96 (dt, J = 15.5, 7.6 Hz, 1H), 5.71 (dd, J = 9.5, 5.0 Hz, 1H), 5.58 (br s, 1H), 5.41 (dd, J = 10.9, 9.4 Hz, 1H), 4.45 (t, J = 8.9 Hz, 1H), 4.24 (dd, J = 9.0, 5.2 Hz, 1H), 4.02 (ddd, J = 13.8, 9.5, 5.0 Hz, 1H), 3.63 (dd, J = 15.7, 8.2 Hz, 1H), 3.48 (dd, J = 15.6, 7.1 Hz, 1H), 2.84–2.68 (m, 1H), 1.98–1.49 (m, 8H); ^{13}C NMR (100 MHz; $CDCl_3$) δ 25.7, 25.8, 29.8, 30.1, 34.4, 43.7, 54.8, 66.2, 69.5, 122.7, 126.4, 128.3, 128.7, 137.0, 139.2, 159.0, 175.7; HRMS (ESI) calcd for $[C_{19}H_{23}NO_4+Na]^+$: 352.1525, found 352.1523.

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