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A Linchpin Synthesis of 6-Hydroxyceramides from Aziridine Aldehydes

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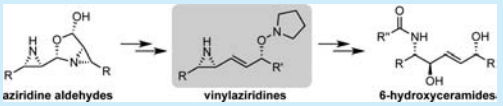
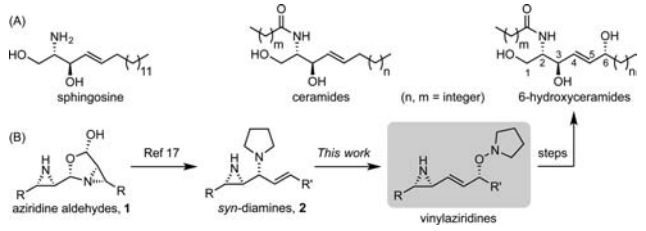
Supporting Information

ABSTRACT: A chemoselective *N*-oxidation/Meisenheimer rearrangement protocol was developed to generate vinylaziridine scaffolds from aziridine aldehydes. A subsequent Lewis acid-mediated aziridine ring opening with carboxylic acid nucleophiles followed by N−O bond cleavage furnishes a human skin 6-hydroxyceramide natural product in short order. The utility of this methodology is demonstrated by the preparation of a number of unnatural 6-hydroxyceramide analogues. This modular approach enables the expedient synthesis of poorly understood skin lipids, which may find application in therapeutics and cosmetics.



Ceramides are sphingosine-based lipids that are crucial for regulating cellular response and are also vital components in the stratum corneum of the human epidermis (Scheme 1A).[1−5] Depending on the appended functional groups, skin ceramides fall into one of two general categories: non-covalently bound extractable lipids and protein-bound lipids. As components in the human body's first line of defense, ceramides have been well-documented in the realm of cosmetics and, more importantly, in studies of dermatitis, melanoma, and cell signaling.[1−5]

Scheme 1. (A) General Molecular Structures of Sphingosine, Ceramides, and 6-Hydroxyceramides; (B) Our Route toward 6-Hydroxyceramides



In the 1990s, a number of new human skin 6-hydroxyceramides were isolated and characterized, and accounted for approximately 30% of the total stratum corneum ceramides.[6−11] To date, the physiological role and biosynthesis of 6-hydroxyceramides are unknown, partially because of the lack of their commercial availability, which hinges on reliable synthetic sequences.[11] To date, five syntheses of 6-hydroxyceramides and their sphingenine core have been reported. In four of these syntheses, the terminal 1,2-amino alcohol moiety was installed via a facially selective attack of the corresponding lithiated alkyne onto Garner's aldehyde.[12−15] Alkene metathesis was also used as the key step to construct the 6-hydroxyceramide core.[16] In addition to long synthetic sequences and the extensive use of protecting groups, these approaches are not amenable to late-stage structural variation. The development of a modular synthesis would not only aid in elucidating the role of 6-hydroxyceramides but also contribute to the development of cosmetics and therapeutics. Herein we present the synthesis of 6-hydroxyceramides from substituted vinylaziridine scaffolds derived from the *syn*-selective borono-Mannich reaction of aziridine aldehyde dimers.[17−19] The vinylaziridine building blocks were subsequently applied to a modular total synthesis of a 6-hydroxyceramide natural product and its unnatural analogues via a series of regio- and chemoselective transformations with minimal reliance on protecting groups (Scheme 1B).

We recently reported a diastereoselective synthesis of *syn*-diamines from aziridine aldehyde dimers.[17] These diamines contain a number of reactive sites that can be employed in downstream modification. Since our studies utilizing diamines **2** as electrophiles for ring opening, we aimed to expand their repertoire by exploring their reactivity as nucleophiles. We sought to selectively distinguish between the two amine centers in **2a** while leaving the alkene untouched by using *N*-oxidation chemistry (Scheme 2). When **2a** was reacted with *m*-CPBA, quantitative *N*-oxidation of the tertiary amine was observed, while the aziridine was left intact. During silica gel chromatography, *N*-oxide **3a** underwent a [2,3]-Meisenheimer rearrangement[20,21] with retention of stereochemistry, exclusively yielding diastereopure γ-aminohydroxy-*trans*-vinylaziridine **4a**. When the reaction was prolonged to 3 days, we observed incomplete conversion to the rearranged product along with a byproduct that resulted from regioselective ring opening of the vinylaziridine with in situ-generated *m*-chlorobenzoic acid (*m*-CBA) (Scheme 2). The transient amino ester **5a** proceeded through an intramolecular *O*- to *N*-acyl transfer to produce byproduct **6a**.

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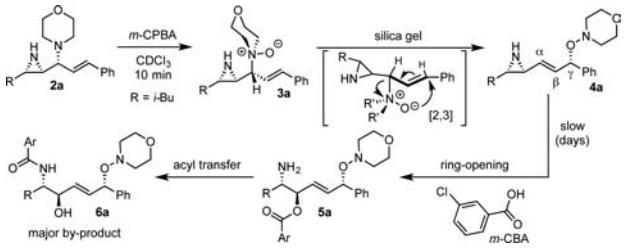
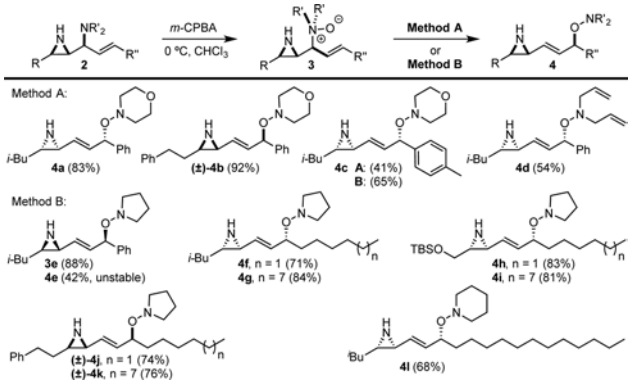
Scheme 2. *N*-Oxidation of Diamine 2a Followed by Rearrangement and Ring Opening



Performing silica gel column purification immediately after oxidation circumvented decomposition. Diastereopure vinylaziridine **4a** was isolated in 83% yield (Scheme 3). The

Scheme 3. Synthesis of Vinylaziridines[a]



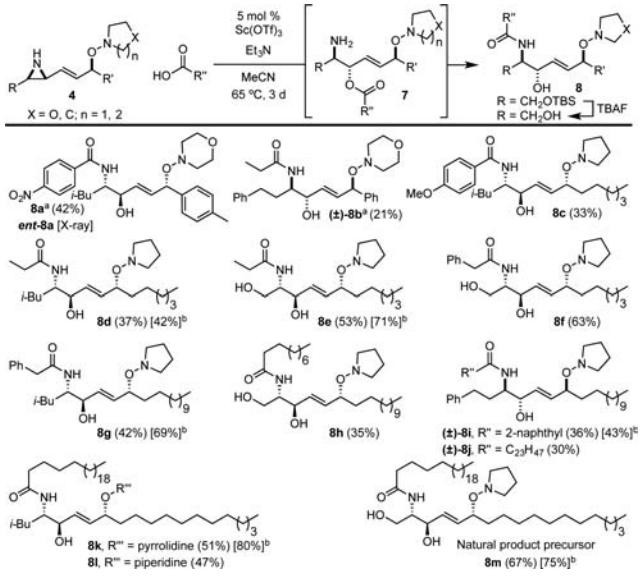
[a]General procedure: 1 equiv of **2** in $CHCl_3$ (0.1 M) at 0 °C followed by 1.1 equiv of *m*-CPBA stirred for 10 min. Method A: the solution was loaded onto silica gel, and the rearranged product eluted. Method B: (i) purification of *N*-oxide followed by stirring in THF for 12 h or (ii) washing with $NaHCO_3$(aq), stirring for 12 h, and then chromatography.

rearrangement was shown to occur readily with various morpholine-derived diamine substrates to produce **4a**–**c** as well as diallylamine **4d**, all of which showed no sign of alkene epoxidation or aziridine ring opening. Tolyl-substituted **4c** was found to rearrange only partially during purification. In addition, when the tertiary amine was derived from pyrrolidine, the [2,3]-rearrangement did not proceed. Only *N*-oxide **3e** was detected by crude ^{1}H NMR analysis and was isolable after elution through a short silica gel column. Fortunately, either stirring the column-purified *N*-oxide in THF or performing a basic workup followed by stirring of the organic phase for 12 h furnished the desired vinylaziridine. Although pyrrolidine-derived **4e** was isolable, it readily decomposed. The rearrangement of tolyl-substituted **4c** was repeated via isolation of the *N*-oxide in an improved yield of 65%.

In order to pursue the total synthesis of 6-hydroxyceramides, new diamine substrates derived from aliphatic alkenyl boronic acids were synthesized using our reported procedure.[17] The multicomponent reaction with the aliphatic alkenyl boronic acids required extended reaction times (24–48 h), and only pyrrolidine and piperidine were found to be suitable secondary amines, as morpholine led to major solvolysis byproducts in HFIP. The diastereomerically pure aliphatic diamines were reacted under the optimized oxidation/rearrangement sequence to produce *trans*-vinylaziridines **4f**–**l** exclusively in good yields (Scheme 3). Overall, the synthesis of *trans*-vinylaziridines with stereochemistry at the γ-position is not easily achieved through conventional methods, especially with the hydroxylamine moiety.[22] These vinylaziridine building blocks are rich in reactive functionality and can be applied to the construction of more complex natural-product-like scaffolds. With the successful chemoselective engagement of the tertiary amine of the diamine in oxidation chemistry, we subsequently aimed to explore the applicability of the γ-chiral vinylaziridines in a concise synthesis of the 6-hydroxyceramide core.

Our success with regioselective ring opening of diamines **2** using *p*-nitrobenzoic acid[17] prompted us to study vinylaziridine ring opening with carboxylic acids en route to the ceramide core. To support our cause, the observation of the initially undesired *m*-chlorobenzoic acid ring opening product **6a** from the *m*-CPBA oxidation indicated that the more electrophilic allylic position of the unactivated vinylaziridine was preferentially attacked. With *O*- to *N*-acyl transfer occurring spontaneously, the 3-hydroxy functional group as well as the new amide bond are generated to complete the core stereocenters of 6-hydroxyceramides without the need for additional chemical steps and protecting groups. Vinylaziridine substrates containing an aryl group at the γ-position (**4a**, **4b**) were initially ring opened in the absence of an activating agent but failed to achieve full conversion when reacted over 4 days. Substrate decomposition and oligomerization were observed at elevated temperatures and in the presence of Lewis acids, respectively. The ring opened products were still isolable from the reaction mixture (**8a**, **8b**), and the regioselectivity was elucidated by 2D NMR correlations (Scheme 4). The absolute stereochemistry was unambiguously confirmed by single-crystal X-ray analysis of *ent*-**8a** (see the Supporting Information (SI)). The molecular structure confirmed not only the facial selectivity of the Meisenheimer rearrangement but also that the ring opening proceeded via an S_N2 reaction.

Scheme 4. Ring Opening of Vinylaziridines[c]



[a]See the SI for the procedure. [b]The assay yield is shown in square brackets. [c]General procedure: vinylaziridine **4** (1 equiv), carboxylic acid (1.5 equiv), and scandium triflate (0.05 equiv) were added to a vial, followed by MeCN (0.05 M) and triethylamine (1.5 equiv), and the mixture was stirred at 65 °C for 72 h.

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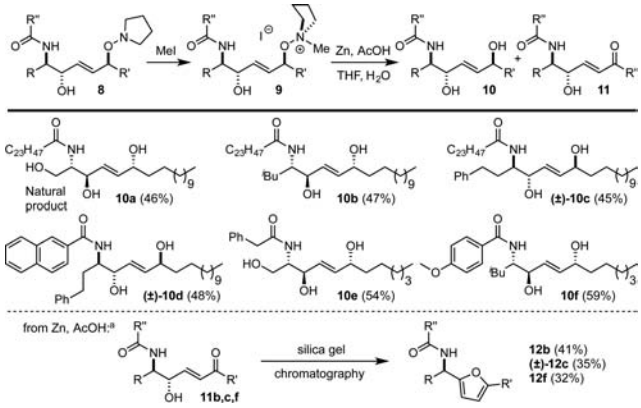
Aliphatic-chain-substituted vinylaziridines were much less reactive toward ring opening in the absence of catalyst and led to low conversion and selectivity with extended reaction times at elevated temperatures. To accelerate the rate of ring opening, we used 10 mol % scandium triflate in the reaction, which furnished the desired product with full conversion of starting material in acetonitrile. The use of $CHCl_3$, THF, or DME as the solvent led to unselective ring opening or decomposition. The catalyst loading substantially influenced the conversion and selectivity of the reaction. We initially employed a catalyst loading of 10 mol % for the ring openings but found that 5 mol % under more dilute conditions (0.05 M) led to a much cleaner reaction by crude ^{1}H NMR analysis (see the SI). We suspect that with higher catalyst loading and concentrations, oligomerization of the transient amino ester **7** with another equivalent of Lewis acid-activated aziridine is more likely to occur (Scheme 4). When the catalyst loading was lowered to 1 or 0.1 mol %, the conversion was comparable to that in the absence of catalyst. The use of 4 Å molecular sieves as a dehydrant led to complete decomposition. In addition, we determined that the presence of triethylamine as a proton shuttle was crucial for conversion to the final ring opened product. On the basis of ^{1}H NMR data, we established that in the presence or absence of Lewis acid, the ring opening reaction proceeds with inversion of stereochemistry.

With these optimized conditions in hand, a series of 6-aminohydroxyceramide analogues were synthesized through ring opening of our *NH*-vinylaziridines with aliphatic and aryl carboxylic acid nucleophiles (Scheme 4). The modest yields of the products do not hinder the overall synthesis, since the N−O bond cleavage would lead immediately to the desired natural product core. The TBS-protected vinylaziridine substrates, in general, were found to furnish the ring opened products in higher yields (**8e**−**f**, **8h**, and **8m**). In all cases, partial TBS deprotection was observed as a result of an undesired scandium triflate-catalyzed reaction in the presence of adventitious water.[23] The mixture could be fully deprotected with TBAF in the same pot simply by changing the solvent to THF. Natural product precursor **8m** was isolated in 67% yield. Conventionally, the ring opening of vinylaziridines with oxygen-based nucleophiles requires the use of strong acids[24−27] or substrate activation via *N*-acylation, Boc protection,[28] benzylation,[29] or sulfonylation.[30−32] In our hands, the use of poorly nucleophilic carboxylic acids under mild Lewis acid catalysis allows for regioselective ring opening of *NH*-vinylaziridines followed immediately by intramolecular acyl transfer; these conditions eliminate the need for additional protecting groups. Given the wide commercial availability of carboxylic acids, there are many possibilities to further understand the function of the amide substituent in the 6-hydroxyceramide natural products through structure−activity relationship studies.

The final step toward the total synthesis of a 6-hydroxyceramide natural product and its analogues was cleavage of the N−O bond to reveal the free 6-hydroxyl group. A number of common conditions for N−O bond cleavage were attempted, such as zinc in acetic acid,[33−36] $Mo(CO)_6$,[37,38] hydrogenolysis,[39,40] samarium diiodide,[41−44] and sodium naphthalenide, all of which were unreactive or led to alkene reduction in the case of hydrogenolysis. According to the literature, N−O bond cleavage is facilitated by the presence of *N*-aryl or *N*-acyl functional groups.[45,46] In the absence of the activated amine, the N−O bond becomes much more difficult, if not impossible, to cleave chemoselectively.[47] Since nucleophilic secondary amines were required for the synthesis of our diamine building blocks, we were unable to use amides and anilines in the multicomponent reaction. Thus, an alternative activation method was required. We envisioned that quaternization of the hydroxylamine nitrogen would be sufficient to activate the N−O bond.[48,49] Methyl iodide was employed at up to 10 equiv in refluxing solvent but did not lead to full conversion. Fortunately, stirring the starting material in neat methyl iodide at room temperature produced the desired ammonium iodide salt **9** with no sign of overmethylation (Scheme 5). With the crude *N*-methylated

Scheme 5. 6-Hydroxyceramide Synthesis[b]



[a]Acid/silica-mediated furan formation of the ketone byproduct. [b]General procedure: **8** was dissolved in MeI and stirred for 12 h. The volatiles were evaporated, followed by stirring with activated Zn (5.5−10 equiv) in 1:1:1 THF/H_2O/AcOH for 8−20 h.

substrate, we were able to cleave the N−O bond in the same pot with reducing agents such as samarium diiodide, sodium naphthalenide, and zinc in acetic acid. We found that zinc in an acetic acid/water/THF solvent mixture furnished the desired product most efficiently while eliminating the need for an inert atmosphere. The natural product **10a** was synthesized in 46% yield over the two sequential steps with a single byproduct corresponding to the α,β-unsaturated ketone **11**. The spectral data for **10a** were in accordance with literature data (see the SI for details).[15] To showcase the versatility of our methodology, other 6-hydroxyceramide analogues were synthesized in modest yields (**10b**−**f**). Byproduct ketones **11b**, **11c**, and **11f** were isolated as their corresponding furans via acid/silica-mediated cyclization, which accounted for the mass balance of the bond cleavage reaction.[50,51]

In conclusion, we have developed a highly modular method for the synthesis of 6-hydroxyceramides from readily available aziridine aldehyde dimers. The synthesis was achieved in short order using a diastereoselective three-component reaction, chemoselective *N*-oxidation/rearrangement sequence to produce chiral vinylaziridines, Lewis acid-mediated regioselective ring opening of the vinylaziridine, then finally N−O bond cleavage via a selective methylation strategy. Our new synthetic sequence is highly tunable, such that any alkenyl boronic acid can be employed with any substituted aziridine aldehyde in the multicomponent reaction and any carboxylic acid may be used in the ring opening step. The high degree of variability in our synthetic method is attractive because of the ability to easily generate a library of 6-hydroxyceramide analogues from common precursors. This methodology not only showcases the first complex natural product synthesis from aziridine aldehyde dimers but also has the potential to further our

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understanding of these human skin natural products toward the development of novel cosmetics and therapeutics.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.orglett.6b03067.

Experimental procedures, X-ray data (CCDC no. 1499251), and NMR spectra (PDF)
Crystallographic data for *ent*-**8a** (CIF)

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Author Contributions

The manuscript was written through contributions of all authors. All authors have approved the final version of the manuscript.

Notes

The authors declare no competing financial interest.

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