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Cyclopropyl Containing Fatty Acids as Mechanistic Probes for Cytochromes P450

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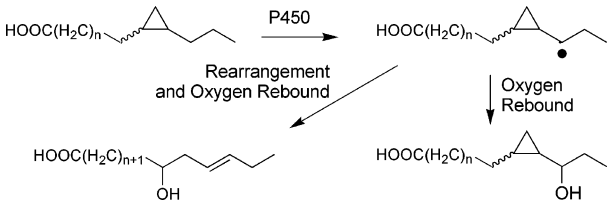
Received November 13, 2004



The mechanism of aliphatic hydroxylation by cytochromes P450 has been the subject of intense debate with several proposed mechanistic alternatives. Various cyclopropyl containing compounds (radical clocks), which can produce both unrearranged and ring opened products upon oxidation, have been key tools in these investigations. In this study, we introduce several cyclopropyl containing fatty acids **1a**−**4a** with which to probe the mechanism of P450s capable of fatty acid hydroxylation. The probes are shown to be capable of distinguishing radical from cationic intermediates due to the rapid equilibration of isomeric cyclopropyl cations. Ring opening of a radical intermediate in an oxidative transformation is expected to yield a single rearranged alcohol, whereas a cation isomerizes prior to ring opening, leading to two isomeric homoallylic alcohols. Oxidation of these probes by $P450_{BM3}$ and $P450_{BioI}$ gives results consistent with a radical but not a cationic intermediate in fatty acid hydroxylation by these enzymes. Quantitation of the unrearranged and ring opened products gives remarkably homogeneous rates for oxygen rebound of $(2-3) \times 10^{10}$ s^{-1}. The effects of introduction of a cyclopropane ring into a fatty acid upon the regiochemistry of hydroxylation are discussed.

Introduction

The cytochromes P450 (P450s) constitute a superfamily of heme-dependent monooxygenases that catalyze a variety of oxidative transformations, including among others, the regio- and stereoselective hydroxylation of aliphatic carbons and carbon−carbon bond cleavage.[1−3] Such reactions play an important role in mammalian xenobiotic detoxification as well as in a wide variety of prokaryotic and eukaryotic biosynthetic and biodegradative pathways. The mechanism of P450 mediated hydroxylation has thus been the subject of intensive investigation for many years. Currently, the accepted mechanistic pathway involves hydrogen atom abstraction by a high valent iron oxo species followed by a rebound of the iron tethered hydroxyl radical onto the substrate carbon radical to give the hydroxylated product.[3] Detection of the substrate radical intermediate predicted by this mechanism provides powerful support for this pathway. One approach, introduced in 1987 and subsequently used extensively for this purpose, employs so-called cyclopropyl radical clocks.[4] These compounds utilize the strain inherent in the cyclopropyl ring system to report the presence of a radical α to the ring via formation of products arising from ring opening reactions. In addition to simply providing evidence for the presence of a radical intermediate, quantitation of both unrearranged and ring-opened oxidation products, coupled with knowledge of the rate of ring opening, allows the lifetime of the radical intermediate to be calculated.

The results from such probes have been generally consistent with the presence of a radical intermediate in

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(1) Ortiz de Montellano, P. R. In *Cytochrome P450: Structure, Mechanism, and Biochemistry,* 2nd ed.; Ortiz de Montellano, P. R., Ed.; Plenum Press: New York, 1995; p 245.

(2) Cryle, M. J.; Stok, J. E.; De Voss, J. J. *Aus. J. Chem.* **2003**, *56*, 749−762.

(3) Ortiz de Montellano, P. R.; De Voss, J. J. *Nat. Prod. Rep.* **2002**, *19*, 477−493.

(4) Ortiz de Montellano, P. R.; Stearns, R. A. *J. Am. Chem. Soc.* **1987**, *109*, 3415−20.

10.1021/jo047985d CCC: $30.25
Published on Web 02/26/2005

the oxidation[4−7] and thus are in harmony with a variety of other experimentally observed features of P450 catalyzed hydroxylations, such as allylic transposition and stereochemical scrambling. However, as increasingly sophisticated probes have been employed to characterize more fully the nature and lifetime of the intermediate, conflicting results have arisen.[8−12] These include calculated lifetimes of radical intermediates more consistent with a transition state and an effectively concerted reaction, as well as indications of a cationic oxidation pathway.[9,11]

Recent density functional theory calculations have reconciled the conflicting results from such probes by suggesting that a two state reactivity paradigm applies to the reactive iron oxo species that mediates P450 oxidations.[13,14] In this, the oxidation can be initiated by one of two different electromers of the iron oxo intermediate, and the reaction then proceeds via two parallel H atom abstraction pathways, one with a true radical intermediate and one that is effectively concerted. Importantly, these studies suggest that, in part, the nature of the substrate may determine which of the two pathways predominates, raising the possibility that sophisticated probes may reveal mechanistic characteristics of their own oxidation and not of all P450 oxidation reactions. In general, the probes employed to date have previously been chosen for, or designed to possess, specific properties, such as faster rates of ring opening or differing ring scission pathways for radical versus cationic intermediates. We thus set out to synthesize a series cyclopropyl radical clocks that, while still reporting on a radical intermediate, were as close as possible to natural P450 substrates.

Our interest in fatty acid metabolizing P450s has led us to investigate $P450_{BM3}$ (CYP102A1), $P450_{BioI}$ (CYP107H1), and CYP119, all of which possess interesting properties. $P450_{BM3}$, a catalytically self-sufficient P450 isolated from *Bacillus megaterium*, possesses one of the fastest rates of turnover for any P450 and is also very well coupled for the hydroxylation of fatty acids and epoxidation of unsaturated acids.[15,16] $P450_{BioI}$, encoded within the biotin operon of *B. subtilis*,[17] is also capable of nonterminal aliphatic hydroxylation.[18] Biosynthetically, however, $P450_{BioI}$ performs carbon−carbon bond cleavage of an unactivated fatty acid chain via alcohol and vicinal diol intermediates.[19,20] The true enzymic substrate is believed to be the acyl carrier protein bound, rather than free, fatty acid. Finally, CYP119 is a P450 isolated from the thermophilic bacteria *Sulfolobus solfataricus* that remains catalytically active at temperatures in excess of 70 °C and is capable of high-temperature hydroxylation of medium chain length fatty acids.[21−23] With the variety of properties that these enzymes possess, it appeared that cyclopropyl radical probes based upon a fatty acid template would be ideal as they would allow us to determine if the mechanism of oxidation in these very different P450s was the same or different in each case. Thus, we report here the synthesis and characterization of a number of novel cyclopropyl fatty acids and demonstrate that they are also capable of distinguishing radical from cationic intermediates. The results of our investigations into the nature of the reactive intermediate in the oxidations catalyzed by these three enzymes employing these probes will also be detailed. A preliminary report of the utility of some these probes with $P450_{BM3}$ has appeared.[24]

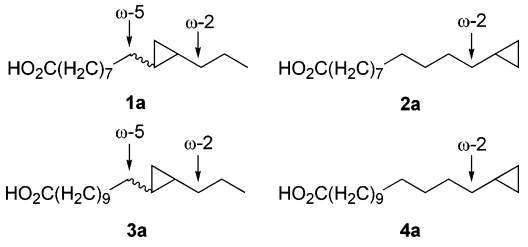


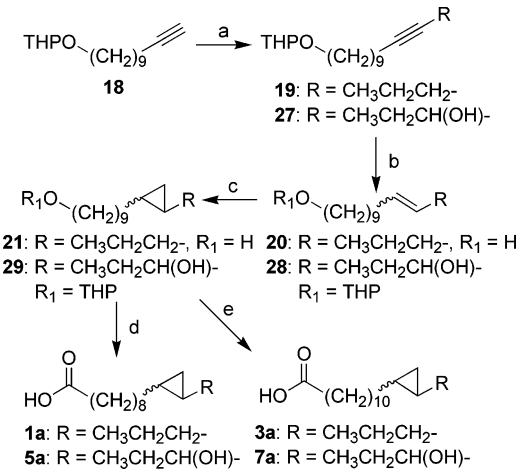
FIGURE 1. Cyclopropyl fatty acids **1a**−**4a** and sites of oxidation for which they would provide mechanistic information.

Results and Discussion

Probe Design and Synthesis. For the purpose of this study, all the probes were designed on a basic fatty acid backbone to exploit ω-2 hydroxylation, which is observed in straight chain fatty acids with $P450_{BM3}$, $P450_{BioI}$, and CYP119 (Figure 1). As such, this allowed the development of three probes for both C_{14} and C_{16} equivalent[25] series of fatty acid probes: these probes possess either a terminal cyclopropyl ring or a *Z* or *E* internal cyclopropyl ring. Such internal cyclopropyl probes would also be able to exploit the ω-5 oxidation site with $P450_{BioI}$ due to the poor regioselectivity of oxidation observed with this

(5) Auclair, K.; Hu, Z.; Little, D. M.; Ortiz de Montellano, P. R.; Groves, J. T. *J. Am. Chem. Soc.* **2002**, *124*, 6020−6027.
(6) Bowry, V. W.; Ingold, K. U. *J. Am. Chem. Soc.* **1991**, *113*, 5699−707.
(7) Bowry, V. W.; Lusztyk, J.; Ingold, K. U. *J. Am. Chem. Soc.* **1991**, *113*, 5687−98.
(8) Newcomb, M.; Johnson, C. C.; Manek, M. B.; Varick, T. R. *J. Am. Chem. Soc.* **1992**, *114*, 10915−10921.
(9) Newcomb, M.; Shen, R.; Choi, S.-Y.; Toy, P. H.; Hollenberg, P. F.; Vaz, A. D. N.; Coon, M. J. *J. Am. Chem. Soc.* **2000**, *122*, 2677−2686.
(10) Newcomb, M.; Shen, R.; Lu, Y.; Coon, M. J.; Hollenberg, P. F.; Kopp, D. A.; Lippard, S. J. *J. Am. Chem. Soc.* **2002**, *124*, 6879−6886.
(11) Toy, P. H.; Newcomb, M.; Hollenberg, P. F. *J. Am. Chem. Soc.* **1998**, *120*, 7719−7729.
(12) Atkinson, J. K.; Ingold, K. U. *Biochemistry* **1993**, *32*, 9209−9214.
(13) Shaik, S.; de Visser, S. P.; Ogliaro, F.; Schwarz, H.; Schroeder, D. *Curr. Opin. Chem. Biol.* **2002**, *6*, 556−567.
(14) Harris, N.; Cohen, S.; Filatov, M.; Ogliaro, F.; Shaik, S. *Angew. Chem., Int. Ed.* **2000**, *39*, 2003−2007.
(15) Boddupalli, S. S.; Pramanik, B. C.; Slaughter, C. A.; Estabrook, R. W.; Peterson, J. A. *Arch. Biochem. Biophys.* **1992**, *292*, 20−28.
(16) Capdevila, J. H.; Wei, S.; Helvig, C.; Falck, J. R.; Belosludtsev, Y.; Truan, G.; Graham-Lorence, S. E.; Peterson, J. A. *J. Biol. Chem.* **1996**, *271*, 22663−22671.
(17) Bower, S.; Perkins, J. B.; Yocum, R. R.; Howitt, C. L.; Rahaim, P.; Pero, J. *J. Bacteriol.* **1996**, *178*, 4122−4130.
(18) Cryle, M. J.; Matovic, N. J.; De Voss, J. J. *Org. Lett.* **2003**, *5*, 3341−3344.
(19) Stok, J. E.; De Voss, J. J. *Arch. Biochem. Biophys.* **2000**, *384*, 351−360.
(20) Cryle, M. J.; De Voss, J. J. *Chem. Commun.* **2004**, 86−87.
(21) Puchkaev, A. V.; Wakagi, T.; Ortiz de Montellano, P. R. *J. Am. Chem. Soc.* **2002**, *124*, 12682−12683.
(22) Koo, L. S.; Immoos, C. E.; Cohen, M. S.; Farmer, P. J.; Ortiz de Montellano, P. R. *J. Am. Chem. Soc.* **2002**, *124*, 5684−5691.
(23) McLean, M. A.; Maves, S. A.; Weiss, K. E.; Krepich, S.; Sligar, S. G. *Biochem. Biophys. Res. Commun.* **1998**, *252*, 166−172.
(24) Cryle, M. J.; Stuthe, J. M. U.; Ortiz de Montellano, P. R.; De Voss, J. J. *Chem. Commun.* **2004**, 512−513.
(25) The probes are actually C_{15} or C_{17} compounds but are considered as derivatives of C_{14} or C_{16} fatty acids as this represents the longest carbon chain in the molecule.

SCHEME 1. Synthesis of the Cyclopropyl Fatty Acids 1a and 3a and the ω-2 Hydroxycyclopropyl Fatty Acids 5a and 6a[a]



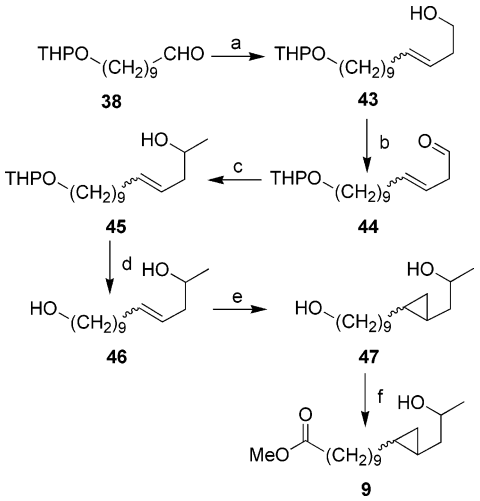
[a] Reagents and conditions: for R = $CH_3CH_2CH_2$- series: (a) *n*-BuLi, THF, HMPA −40 °C, then $CH_3CH_2CH_2Br$, 81%. (b) (i) *p*-TsOH, MeOH; (ii) *Z*: H_2, Lindlar catalyst, hexanes, 86%; *E*: Li, NH_3, *t*-BuOH, THF, −40 °C, 68%. (c) CH_2I_2, Et_2Zn, TFA, CH_2Cl_2, 0 °C, 97−100%. (d) CrO_3, H_2SO_4, Me_2CO, 0 °C, 70−91%. (e) (i) $(COCl)_2$, DMSO, CH_2Cl_2, −78 °C, then TEA; (ii) Ph_3PCHCO_2Me, $CHCl_3$, reflux; (iii) H_2, PtO, hexanes; (iv) LiOH, MeOH, 10−15% (four steps). For R = $CH_3CH_2CH(OH)$- series: (a) *n*-BuLi, THF, −40 °C, then EtCHO, 99%. (b) *Z*: H_2, Lindlar catalyst, hexanes, 100%; *E*: LAH, THF, 0 °C, 99%. (c) CH_2I_2, Et_2Zn, TFA, CH_2Cl_2, 0 °C, 98−100%. (d) (i) *p*-TsOH, MeOH; (ii) CrO_3, H_2SO_4, acetone, 0 °C; (iii) $NaBH_4$, $CeCl_3$, MeOH, 0 °C, 69−83% (three steps). (e) (i) $(COCl)_2$, DMSO, CH_2Cl_2, −78 °C, then TEA; (ii) Ph_3PCHCO_2Me, $CHCl_3$, reflux; (iii) $NaBH_4$, $CeCl_3$, MeOH, 0 °C; (iv) H_2, PtO, hexanes; (v) LiOH, MeOH, 17−19% (five steps).

enzyme.[18] While this work was ongoing, a report appeared detailing the synthesis and unsuccessful use of **3a**[26] and other related compounds as probes of the mechanism of a non-heme iron desaturase.[27] In this case, although enzymic products were observed, no rearrangement products were detected.

The terminal acetylene **18** was considered a key precursor in the expedient synthesis of both the disubstituted cyclopropanes **1a** and **3a** and their expected oxidation products **5a** and **7a**, respectively (Scheme 1). This was indeed the case as alkylation of **18** cleanly provided the internal alkyne **19**, which could be converted selectively to the corresponding *Z* or *E* alkene **20** by reduction with either hydrogen in the presence of Lindlar's catalyst (>98% *Z*) or lithium in liquid ammonia (>95% *E*). Simmons−Smith cyclopropanation[28] of **20** and oxidation of the alcohol terminus led directly to the C_{14} probes **1a**, while the C_{16} probes **3a** required a two carbon chain extension via a Wittig condensation reaction.

An analogous route was used to synthesize products **5a** and **7a** expected from P450 mediated oxidation of **1a** and **3a** except that it was initiated by addition of **18** to the carbonyl of propanal. Fortuitously, as the sodium borohydride reduction of cyclopropyl ketones, the method

SCHEME 2. Synthesis of the ω-1 Hydroxylated Cyclopropyl Fatty Acid Ester 9[a]



[a] Reagents and conditions: (a) $Ph_3P(CH_2)_3OH$, *n*-BuLi, Et_2O, −78 °C, 86%. (b) PCC, NaAc, CH_2Cl_2, 94%. (c) $BrMgCH_3$, Et_2O, 87%. (d) *p*-TsOH, MeOH, 91%. (e) CH_2I_2, Et_2Zn, TFA, CH_2Cl_2, 0 °C, 89%. (f) (i) CrO_3, H_2SO_4, Me_2CO, 0 °C; (ii) CH_2N_2, Et_2O; (iii) $NaBH_4$, $CeCl_3$, MeOH, 0 °C, 90% (two steps).

chosen to install the hydroxyl moiety, is known to proceed with a strong preference for production of the anti diastereomer,[29] we were also able to use the products **5** and **7** to establish the diastereoselectivity of the P450 mediated oxidation.

The other likely unrearranged products from P450 mediated oxidation of **1a** and **3a** correspond to ω-1 oxidation. The aldehyde **38**, available via ozonolysis of protected undec-11-en-1ol, provided the differentially protected terminal diol **43** as a 1:1 mixture of *E* and *Z* isomers after Wittig chain extension (Scheme 2). Methyl Grignard addition to the corresponding aldehyde, cyclopropyl ring formation, and manipulation of the oxidation states of the oxygenated carbons provided the hydroxy ester **9** as an equal mixture of all four diastereomers. As expected, none of the distereoselectivity seen in the reduction of the α-cyclopropyl ketones precursors to **5a** and **7a** was observed in this series.

Oxidative rearrangement of the disubstituted cyclopropanes **1a** and **3a** could yield either a primary (**11a**, **12a**) or a secondary (**13a**, **14a**) alcohol, depending on the ring scission pathway followed (Figure 2). The products from the C_{14} series were prepared as primary standards as well as to provide an indication of GCMS fragmentation patterns for the C_{16} series.

The acid **13a** is the most likely product of oxidative rearrangement of **1a** as the rate of ring opening of the corresponding cyclopropyl carbinyl radical to afford a secondary radical (Figure 2, path b) is faster than the alternative fragmentation that leads to **11a** (Figure 2, path a). Synthesis of **13** was achieved via allylmagnesium bromide addition to the aldehyde **38**, which in turn was derived from ozonolysis of THP-protected undec-10-en-1-ol (Scheme 3). Subsequent ozonolysis of **39** and Wittig chain extension gave the desired 15-carbon chain in **40** with the required unsaturation and oxygenation. Func-

(26) If an acid and the corresponding methyl ester are discussed in this paper, a compound number followed by an "a" indicates the acid, and the compound number alone indicates the ester.

(27) Villorbina, G.; Roura, L.; Camps, F.; Joglar, J.; Fabrias, G. *J. Org. Chem.* **2003**, *68*, 2820−2829.

(28) Zhiqiang, Y.; Lorenz, J. C.; Shi, Y. *Tetrahedron Lett.* **1998**, *39*, 8621−8624.

(29) Kazuta, Y.; Abe, H.; Yamamoto, T.; Matsuda, A.; Shuto, S. *J. Org. Chem.* **2003**, *68*, 3511−3521.

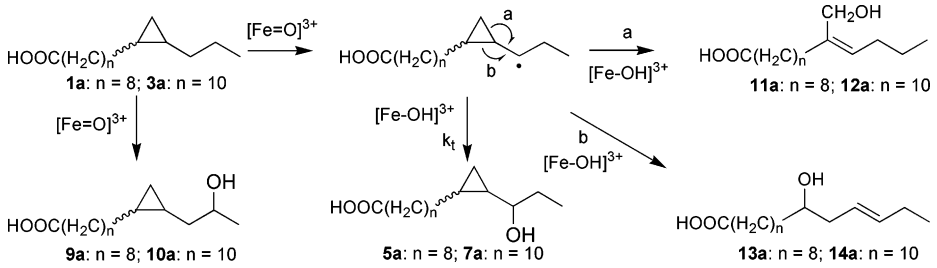
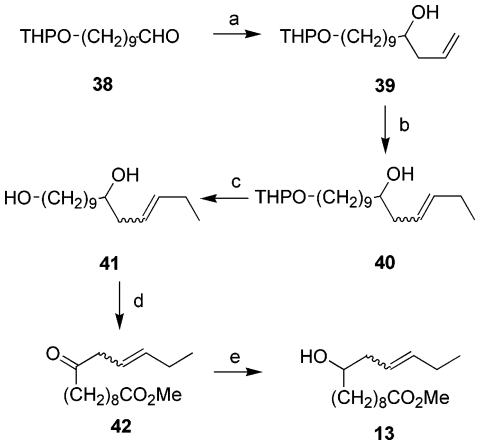


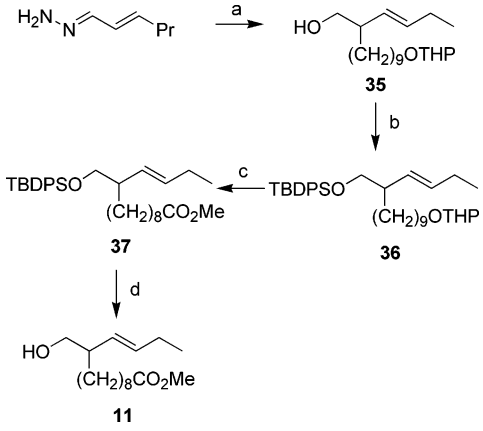
FIGURE 2. Possible products from the P450 mediated hydroxylation of cyclopropyl fatty acids **1a** and **3a**.

SCHEME 3. Synthesis of the Unsaturated Hydroxyester 13[a]



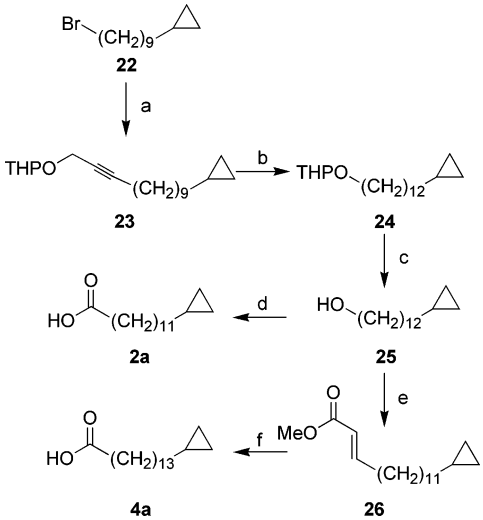
[a] Reagents and conditions: (a) AllylMgBr, Et_2O, −40 °C, 87% (b) (i) O_3, CH_2Cl_2, −78 °C then DMS; (ii) $Ph_3P(CH_2)_2CH_3$, *n*-BuLi, Et_2O, −78 °C, 72% (two steps). (c) *p*-TsOH, MeOH, 90%. (d) (i) CrO_3, H_2SO_4, Me_2CO, 0 °C; (ii) CH_2N_2, Et_2O, 88%. (e) $NaBH_4$, MeOH, 0 °C, 75%.

SCHEME 4. Synthesis of the Unsaturated Hydroxy Ester 11[a]



[a] Reagents and conditions: (a) (i) LDA, THF, −78 °C, then $Br(CH_2)_9OTHP$; (ii) $CuCl_2$; (iii) $NaBH_4$, MeOH, 0 °C, 20% (three steps). (b) TBDPSCl, imidazole, CH_3CN, 65%; (c) (i) *p*-TsOH, MeOH; (ii) CrO_3, H_2SO_4, Me_2CO, 0 °C; (iii) CH_2N_2, Et_2O, 80% (three steps). (d) TBAF, THF, 100%.

SCHEME 5. Synthesis of the Cyclopropyl Fatty Acids 2a and 4a[a]



[a] Reagents and conditions: (a) (i) 1-(tetrahydropyran-2′-yloxy)-prop-2-yne, *n*-BuLi, THF, HMPA, −40 °C, then **22**, 85%. (b) H_2, Pd/C, hexanes, 99%. (c) *p*-TsOH, MeOH, 98%. (d) CrO_3, H_2SO_4, Me_2CO, 0 °C, 89%. (e) (i) PCC, CH_2Cl_2; (ii) Ph_3PCHCO_2Me, $CHCl_3$, reflux, 57% (two steps). (f) (i) H_2, Pd/C, hexanes; (ii) LiOH, MeOH, 64% (two steps).

tional group manipulation of **40** then gave **13** as a mixture of *E*/*Z* isomers.

The rearrangement product **11a** would be formed from ring opening of the cyclopropylcarbinyl radical derived from **1a** to afford a primary radical (Figure 2). Alkylation of the α,*β*-unsaturated dimethyl hydrazone derived from hex-2-enal with THP protected 9-bromononan-1-ol to yield a *β*,*γ*-unsaturated alcohol **35** after reduction was the key step in the synthesis of **11** (Scheme 4).[30,31] Manipulation of protecting groups and adjustment of the oxidation state of **35** provided the desired ester **11**.

The monosubstituted cyclopropanes **2a** and **4a** were synthesized via initial alkylation of THP protected propargyl alcohol with a 12-carbon *ω*-cyclopropyl bromide **22** (available in two steps from commercial undec-10-en-1-ol) (Scheme 5). Following relevant functional group manipulation, this afforded **2a** and then **4a** using Wittig chain-extension methodology.

Ozonolysis of cyclododecene employing Schreiber's[32] methodology produced differentiated, terminally difunctionalized C_{12} compounds that allowed access to both unrearranged and rearranged oxidation products arising from **2a** and **4a** (Figure 3). Thus, acetal aldehyde **30** smoothly undergoes vinyl Grignard addition and cyclopropanation as the key steps in the synthesis of **6a** and **8a** (Scheme 6). The product expected from oxidative

(30) Mino, T.; Fukui, S.; Yamashita, M. *J. Org. Chem.* **1997**, *62*, 734−735.
(31) Yamashita, M.; Matsumiya, K.; Nakano, K. *Bull. Chem. Soc. Jpn.* **1993**, *66*, 1759−1763.
(32) Schreiber, S. L.; Claus, R. E.; Reagan, J. *Tetrahedron Lett.* **1982**, *23*, 3867−3870.

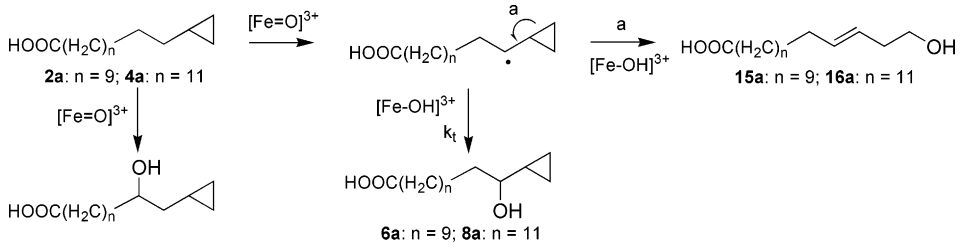
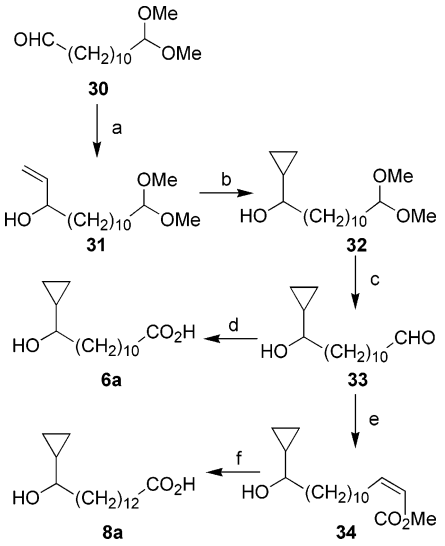


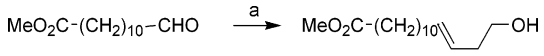
FIGURE 3. Possible products from the P450 mediated hydroxylation of cyclopropyl fatty acids **2a** and **4a**.

SCHEME 6. Synthesis of the ω-2 Hydroxycyclopropyl Fatty Acids 6a and 8a[a]



[a] Reagents and conditions: (a) $BrMgCH{=}CH_2$, Et_2O, 92%. (b) CH_2I_2, Et_2Zn, TFA, CH_2Cl_2, 0 °C, 92%. (c) *p*-TsOH, Me_2CO, 97%. (d) (i) CrO_3, H_2SO_4, Me_2CO, 0 °C; (ii) $NaBH_4$, $CeCl_3$, MeOH, 0 °C, 62%. (e) Ph_3PCHCO_2Me, $CHCl_3$, reflux, 79%. (f) (i) H_2, Pd/C, hexane; (ii) LiOH, MeOH, 55% (two steps).

SCHEME 7. Synthesis of the Unsaturated Hydroxyester 15[a]

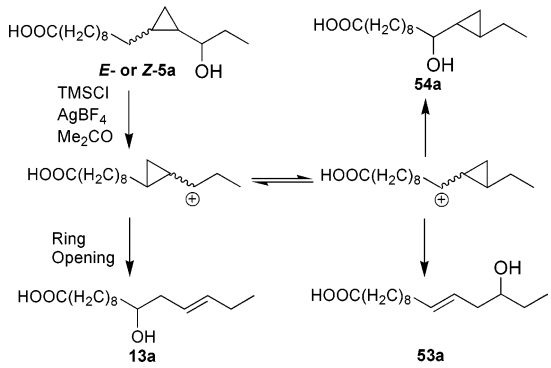


[a] Reagents and conditions: (a) $Ph_3P(CH_2)_3OH$, *n*-BuLi, Et_2O, −78 °C, 60%.

rearrangement of **2a** is **15a** (Figure 3), and the corresponding methyl ester standard **15** was afforded in one step via a selective Wittig condensation with methyl 12-oxododecanoate **48**, also available via ozonolysis of cyclododecene[32] (Scheme 7).

Chemical Rearrangement of Cyclopropyl Cation. The products of rearrangement of the cyclopropyl cation derived from **1a** under Lewis acidic conditions were investigated. It was found that two isomeric homoallylic alcohols and two isomeric cyclopropyl alcohols were present in the reaction mixture, both in a 1:1 ratio. Separation of these two pairs of compounds by chromatography on silver impregnated silica gel after esterification with diazomethane and extensive 2-D NMR experiments identified the homoallylic alcohols as **13** and **53** and the cyclopropyl containing compounds as **1** and **54**. This suggests that the initially formed cation rapidly equilibrates between the two degenerate, stabilized forms

SCHEME 8. Equilibration of Cyclopropyl Cation in the Rearrangement of 5a



(Scheme 8) and more slowly undergoes ring opening to a homoallylic cation that can be quenched to yield the observed unsaturated alcohols. The 1:1 ratio of these homoallylic alcohols mandates a much faster rate of equilibration than of ring opening for the cyclopropyl cations. Such a rapid equilibration between degenerate cations has previously been observed spectroscopically by Olah in superacid solution.[33] Importantly, if a cationic ring opening is proposed as the origin for any homoallylic alcohol **13a** observed in a P450 mediated oxidation of **1a**, it would be expected to be accompanied by an equal amount of the isomeric **53a**, as well as the rearranged cyclopropyl alcohol **54a**. This would analogously hold for the oxidation of **3a**. Although it is possible that the enzyme active site could impose significant restraints upon the molecular motion of any intermediate and thus direct the product distribution, the lack of any cation derived product in both the C_{14} and C_{16} equivalent series makes this an unlikely explanation. In addition, active site constraints have previously been discounted with other cyclopropyl containing P450 substrates.[34] Thus, **1a** and **3a** should not only provide a means of detection of a radical or cationic intermediate in a P450 mediated oxidation, but they should also serve to distinguish between the two.

GCMS Analysis of Cyclopropyl Fatty Acid Esters. GCMS analysis of the methyl esters of the initially produced acids was used as the major method for identification of the structures of the products from P450 mediated oxidation of **1a**−**4a**. A number of fragmentation patterns characteristic of the position of hydroxylation along a saturated fatty acid ester chain have previously been reported by us[18] and others[35−37] (Scheme 9). Generally, major peaks are seen for scission α to the hydroxyl

(33) Olah, G. A.; Reddy, V. P.; Prakash, G. K. S. *Chem. Rev.* **1992**, *92*, 69−95.
(34) Newcomb, M.; Le Tadic, M.-H.; Putt, D. A.; Hollenberg, P. F. *J. Am. Chem. Soc.* **1995**, *117*, 3312−3313.

SCHEME 9. Major MS Fragmentation Pathways of Hydroxy Fatty Acid Methyl Esters

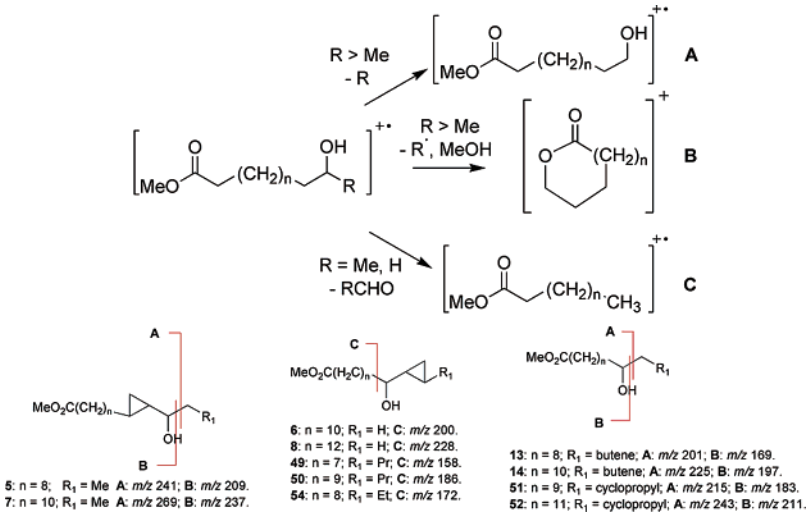


TABLE 1. Regioselectivity of Hydroxylation by $P450_{BM3}$ of C_{14} and C_{16} Straight Chain Fatty Acids and Cyclopropyl Fatty Acids 1a−4a

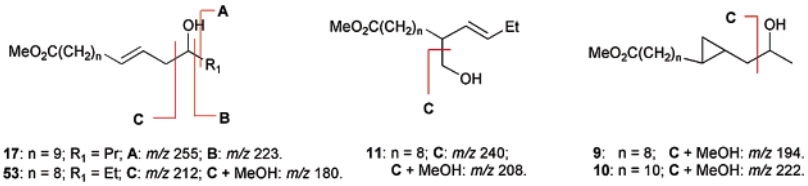
		percent hydroxylation			
probe	coupling[45] (%)	>ω-3 (%)	ω-3 (%)	ω-2 (%)	ω-1 (%)
tetradecanoate	70	0	21 ± 2	32 ± 2	47 ± 3
***Z*-1a**	73	0		33 ± 2 (21:12)[a] 1.0 ± 0.1 **13a**	66 ± 9
***E*-1a**	70	(ω-5) 11 ± 1		70 ± 4 (1:1)[a]	19 ± 1
2a	80	0	14 ± 1	86 ± 4	
hexadecanoate	68	(ω-5) 0.5 (ω-4) 1.5	33 ± 3	43 ± 3	22 ± 2
***Z*-3a**	75	0		48 ± 1 (2:1)[a] 1.5 ± 0.1 **14a**	50 ± 8
***E*-3a**	80	0		100 (syn)[a]	0
4a	55	0	11 ± 1	89 ± 3	

[a] Syn to anti ratio displayed in parentheses.

with resultant loss of the alkyl chain (pathway A, Scheme 9) and lactonization and subsequent α scission (pathway B, Scheme 9). For hydroxy fatty acid esters where the hydroxyl is in the ω or ω-1 position, hydrogen atom transfer and loss of an aldehyde is observed (pathway C, Scheme 9). It was important to clarify whether these major fragmentation patterns were still observed in hydroxy fatty acid esters bearing cyclopropyl (or alkene) moieties near the alcohol group, which was permitted via GCMS analysis of the suite of synthetic standards synthesized previously. The fragmentations of Scheme 9 in the main provided explanations for the fragments seen in these compounds, although the presence of the cyclopropyl could effect partitioning between the A/B or the C pathway in some cases and induce novel pathways in others. Thus, a cyclopropyl α to the hydroxyl and away from the carboxyl terminus of the chain induces fragmentation by pathway C (**6** and **8** Figure 4). Functionality β to the hydroxyl but still distal to the carboxyl moiety has little effect, and paths A/B are observed (**13** and **14** Figure 4). If the cyclopropyl ring is proximal to the carboxyl terminus and α to the hydroxyl paths, A/B operates although it is clear that alternative fragmentation pathways also begin to compete with these (**5** and **7** Figure 4). If the functionality is proximal to the carboxyl but β to the hydroxyl, then the major fragments observed generally arise from a novel pathway, corresponding to loss of methanol and a terminal aldehyde, presumably via directed fragmentation of an intermediate lactone (**9** and **10** Figure 4). These generalizations hold true not only for the synthetic standards but for all of the identified products of enzymic oxidation and chemical rearrangement seen in this work (vide infra) (Figure 4).

Enzyme Catalyzed Oxidation of Cyclopropyl Fatty Acids. All of the synthesized cyclopropyl fatty acids **1a**−**4a** were incubated with $P450_{BM3}$, $P450_{BioI}$, and CYP119 in the presence and absence of NADPH (or NADH), and for $P450_{BioI}$ and CYP119, appropriate electron-transfer partners. $P450_{BM3}$ oxidized all of the fatty acids, $P450_{BioI}$ oxidized all but ***E*-3a**, while CYP119 only gave detectable products with ***Z*-1a**, **2a**, and ***E*-3a**. The products were identified by GCMS analysis, and where appropriate, comparison with synthetic standards and the regiochemistry of oxidation is given in Tables 1−3. The levels of turnover were such that mechanistic information could be derived for only $P450_{BM3}$ and $P450_{BioI}$, but some information on the influence of the cyclopropyl moiety on the regiochemistry of oxidation for all three enzymes

(35) Kawamura, K.; Gagosian, R. B. *J. Chromatogr.* **1988**, *438*, 309−317.
(36) Kerwin, J. L.; Torvik, J. J. *Anal. Biochem.* **1996**, *237*, 56−64.
(37) Wilson, R.; Smith, R.; Wilson, P.; Shepherd, M. J.; Riemersma, R. A. *Anal. Biochem.* **1997**, *248*, 76−85.



17: n = 9; R_1 = Pr; **A**: *m/z* 255; **B**: *m/z* 223. **53**: n = 8; R_1 = Et; **C**: *m/z* 212; **C** + MeOH: *m/z* 180.

11: n = 8; **C**: *m/z* 240; **C** + MeOH: *m/z* 208.

9: n = 8; **C** + MeOH: *m/z* 194. **10**: n = 10; **C** + MeOH: *m/z* 222.

FIGURE 4. Major fragmentation pathways and ions seen in mass spectra of a variety of hydroxy fatty acid esters.

TABLE 2. Regioselectivity of Hydroxylation by $P450_{BioI}$ of C_{14} and C_{16} Straight Chain Fatty Acids and Cyclopropyl Fatty Acids 1a−4a

		percent hydroxylation			
probe	coupling[45] (%)	>ω-3 (%)	ω-3 (%)	ω-2 (%)	ω-1 (%)
tetradecanoate[18]	6	0	49 ± 1	30 ± 1	21 ± 1
***Z*-1a**	8	0		58 ± 1 (37:21)[a] 1.6 ± 0.3 **13a**	41 ± 3
***E*-1a**	7	0		77 ± 7 (2:1)[a]	23 ± 4
2a	6	0	69 ± 11	31 ± 4	
hexadecanoate[18]	2	(ω-5) 12 ± 2 (ω-4) 20 ± 2	24 ± 2	29 ± 2	15 ± 2
***Z*-3a**	13	(ω-5) 73 ± 5 (4:1)[a] **50a** 2.2 ± 0.8 **17a**		16 ± 2 (1:1)[a]	9 ± 2
***E*-3a**	0	0		0	0
4a	<1	0	~90	~10	

[a] Syn to anti ratio displayed in parentheses.

TABLE 3. Regioselectivity of Hydroxylation by CYP119 of C_{14} and C_{16} Straight Chain Fatty Acids and Cyclopropyl Fatty Acids *Z*-1a, 2a, and *E*-3a

	percent hydroxylation			
probe	>ω-3 (%)	ω-3 (%)	ω-2 (%)	ω-1 (%)
tetradecanoate	(ω-5) 13 (ω-4) 66	17	4	0
***Z*-1a**	0	0	100	0
2a	0	~20	~80	

	percent hydroxylation			
probe	ω-7 (%)	ω-6 (%)	ω-5 (%)	ω-4 (%)
hexadecanoate	17	76	7	0
***E*-3a**	0	trace	0	0

is available. The results will be discussed under these two broad categories.

Mechanism of P450—a Radical But Not a Cation Intermediate. The products from the P450 catalyzed oxidation of compounds **1a**−**4a** were carefully examined for both the products of direct hydroxylation α to the cyclopropyl ring and the rearrangement products arising from either radical or cationic intermediates involved in this oxidation. Because of the relative rates of cyclopropyl ring opening,[6,7] the most likely compounds to exhibit rearrangement products were ***Z*-1a** and ***Z*-3a** predicted to ring open 4 times faster than **2a** and **4a** and >3 times faster than ***E*-1a** and ***E*-3a**. Previously, we reported that ***Z*-1a** and ***Z*-3a**, when incubated with $P450_{BM3}$, did indeed produce both the α-hydroxylated products **5a** and **7a** as well as the rearranged products **13a** and **14a**, respectively.[24] None of the other products (**53a** or **54a**) expected from rearrangement of a cationic intermediate were observed, and the ratio of the rearranged to nonrearranged products allowed an upper limited of 2.6×10^{10} s^{-1} to be calculated for the rate of oxygen rebound.[24] We now report that the incubation of ***Z*-1a** and ***Z*-3a** with $P450_{BioI}$ again results in high levels of oxidation α to the cyclopropyl ring, although in the case of ***Z*-3a** this is at the ω-5 position and yields **50a**, whereas for ***Z*-1a** reaction occurs at the ω-2 carbon (**5a**) (Table 2). With $P450_{BM3}$, oxidation only occurred at the ω-1 position in both ***Z*-1a** and ***Z*-3a** (Table 1). Along with the products of direct hydroxylation of ***Z*-1a** and ***Z*-3a**, the homoallylic alcohols expected from rearrangement of an intermediate radical were also observed. No isomeric cyclopropyl or homoallylic alcohols expected from rearrangement of cationic intermediates were detected in any of the enzymic reactions. Quantitation (Table 2) of the α-cyclopropyl alcohol and the corresponding rearranged homoallylic alcohol (***Z*-1/13** and ***Z*-3/50**) and utilization of a ring opening rate of 8.0×10^8 s^{-1} for *Z*-disubstituted cyclopropanes[6,7] allowed calculation of the rebound rate for the Fe(IV)-OH and the carbon radical pair in each case. The rates obtained were in excellent internal agreement (2.9×10^{10} s^{-1} for ***Z*-1a** and 2.7×10^{10} s^{-1} for ***Z*-3a**) with each other and with the results obtained for $P450_{BM3}$ oxidation of the same probes (2.6×10^{10} s^{-1}).

The two state reactivity theory of P450 mechanism suggests that the nature and structure of the substrate may influence partitioning between the nonconcerted and effectively concerted pathways. This has been used to explain the very different rebound rates calculated from

data obtained with different cyclopropyl containing probes.[3] It was thus important to determine whether the structural differences between ***Z*-1a/*Z*-3a** and ***E*-1a/*E*-3a** and the monosubstituted cyclopropanes **2a** and **4a**, which lead to significant differences in the rate of ring opening, led to different apparent rebound rates. No rearranged homoallylic alcohol was detected in the P450 catalyzed oxidation of **2a** or **4a**, but this was to be expected based upon the rebound rate calculated previously, the expected rate of ring opening, and the sensitivity of detection. However, in the oxidation of ***E*-1a** and ***E*-3a** by $P450_{BM3}$, the rearranged homoallylic alcohols **13** and **14** were detectable by analysis of single ion chromatograms following after esterification with diazomethane, corresponding to their major fragment ions. This was necessary as **13** and **14** coeluted under analysis conditions with the major ω-2 oxidation products **5** and **7**, respectively, which unfortunately also precluded accurate quantitation. A large scale oxidation of ***E*-1a** (0.02 mmol) by $P450_{BM3}$ was conducted, the products of turnover were analyzed using 500 MHz ^{1}H NMR, and the peaks corresponding to the vinylic hydrogens of **13** and the methine hydrogens α to both the hydroxyl and the cyclopropyl of **5** were quantified by integration. These imprecise measurements suggested a rebound rate of 2.2×10^{10} s^{-1} (see Supporting Information).

The close correspondence of the rebound rates calculated for ***Z*-1a** and ***Z*-3a** oxidation with both $P450_{BM3}$ and $P450_{BioI}$ and the qualitatively similar results obtained with ***E*-1a**, which undergoes ring opening considerably more slowly, is striking. Rebound rates from the same probe in different enzymes,[5,10] or from very similar probes with the same enzyme,[38] have often been significantly different. Two state reactivity theory explains the significant differences observed by suggesting that the differences in enzyme or substrate structure determine the flux through nonconcerted and effectively concerted pathways and thus alter the observed rebound rate. The results reported in this work suggest that for a matched enzyme substrate pair—both $P450_{BM3}$ and $P450_{BioI}$ naturally utilize fatty acids, and the probes used here closely resemble the natural substrates—the differences in pathway utilization are small.

Effect of Cyclopropyl Moiety on the Regiochemistry of Hydroxylation. The products from $P450_{BM3}$, $P450_{BioI}$, and CYP119 catalyzed oxidation of **1a**−**4a** allowed not only determination of the presence and lifetime of a radical intermediate but also demonstrated the effect a cyclopropyl moiety in the fatty acid chain has upon the regiochemistry of oxidation. $P450_{BM3}$ binds straight chain fatty acids, oxidizes them in a tightly coupled fashion (NADPH consumption: product formation $\approx$1), and is known to catalyze their oxidation (at the ω-1 to ω-3 positions) with moderate regioselectivity and high enantioselectivity.[15,16,39] All of the probes used in this study were oxidized in a highly coupled fashion by $P450_{BM3}$. The cyclopropyl fatty acids ***E*-1a**, ***E*-3a**, **2a**, and **4a** would be expected to have extended, albeit somewhat rigidified, conformations analogous to those seen in straight chain fatty acids. Oxidation of these fatty acids (***E*-1a**, ***E*-3a**, **2a**, and **4a**) exhibited changes in regiochemistry attributable to weakening of the pseudo-allylic C−H bonds α to the cyclopropyl ring.[40] Such electronic effects in determining the regiochemistry of P450 oxidation have been reported previously, with an order of preference for oxidation site in the absence of binding effects paralleling bond strength (i.e., CH > CH_2 > CH_3).[40−43] In fact, in the case of ***E*-3**, only the product of hydroxylation α to the cyclopropyl is observed, an unprecedented regioselectivity for fatty acid hydroxylation by this enzyme. The significant amount of ω-5 oxidation observed with the relatively short ***E*-1a** is similarly unusual in fatty acid hydroxylation by wild-type $P450_{BM3}$. Epoxidation has been found a similar distance from the methyl terminus of arachadonic acid,[16] and very low levels of ω-5 hydroxylation have previously been reported for oxidation of the longer palmitate.[39,44] Collectively, the results demonstrate that a somewhat surprising degree of flexibility must exist for the binding of substrates by $P450_{BM3}$. The fatty acids ***Z*-1a** and ***Z*-3a** would be conformationally distinct from the other fatty acids used in this study as well as straight chain ones because of the *Z* ring geometry, which would be expected to significantly reduce the binding alternatives of the substrates to the enzyme. The interplay of electronic and binding effects in these cases results in a significant amount of oxidation both α to the cyclopropyl in the electronically favored position and at the sterically more accessible β carbon.

$P450_{BioI}$ is known to be much less regioselective and enantioselective than $P450_{BM3}$ in its oxidation of straight chain fatty acids.[18] Moderate enantioselectivity is seen across the ω-1 to ω-3 positions in tetradecanoic acid, while similar enantioselectivity is seen from the ω-1 to ω-5 position in hexadecanoic acid.[18] The lack of regioselectivity of $P450_{BioI}$ is even more striking in that the C7−C8 bond cleavage via a cascade of oxidative steps in tetradecanoic acid is also seen and is believed to be related to its physiological function.[20] Additionally, although fatty acids bind well, their natural substrate is believed to be a fatty acid thioester of an acyl carrier protein.[19] Not surprisingly, therefore, the results of $P450_{BioI}$ catalyzed oxidation of the probes were varied. The disubstituted cyclopropyl fatty acids **1a** and **3a** in general showed increased oxidation at the pseudoallylic positions α to the three membered ring. The C_{14} equivalent acids **1a** and **2a** were better substrates than analogous straight chain tetradecanoic acid and the C_{16} equivalent acids **3a** and **4a**, and in fact, ***E*-3a** was not oxidized at all by $P450_{BioI}$. In the shorter probe **1a**, oxidation was predominantly at the ω-2 position, while in the longer ***Z*-3a** it was at the ω-5 position proximal to the carboxyl terminus. The monosubstituted cyclopropyl fatty acids **2a** and **4a** were oxidized at the presumably more accessible ω-3 position rather than the more reactive ω-2 position α to the cyclopropyl ring.

(38) He, X.; Ortiz de Montellano, P. R. *J. Biol. Chem.* **2004**, *279*, 39479−39484.
(39) Truan, G.; Komandla, M. R.; Falck, J. R.; Peterson, J. A. *Arch. Biochem. Biophys.* **1999**, *366*, 192−198.
(40) de Visser, S. P.; Kumar, D.; Cohen, S.; Shacham, R.; Shaik, S. *J. Am. Chem. Soc.* **2004**, *126*, 8362−8363.
(41) Korzekwa, K. R.; Jones, J. P.; Gillette, J. R. *J. Am. Chem. Soc.* **1990**, *112*, 7042−7046.
(42) White, R. E.; McCarthy, M. B.; Egeberg, K. D.; Sligar, S. G. *Arch. Biochem. Biophys.* **1984**, *228*, 493−502.
(43) Frommer, U.; Ullrich, V.; Staudinger, H. *Hoppe-Seyler's Z. Physiol. Chem.* **1970**, *351*, 903−912.
(44) Murataliev, M. B.; Trinh, L. N.; Moser, L. V.; Bates, R. B.; Feyereisen, R.; Walker, F. A. *Biochemistry* **2004**, *43*, 1771−1780.

CYP119 has previously been reported to bind fatty acids of varying chain length and to oxidize lauric acid with moderate regioselectivity (ω/ω-1/ω-2/ω-3 7:70:22:1).[22] The data obtained in this work for the oxidation of tetradecanoate and hexadecanoate (Table 3) again show only moderate regioselectivity. Interestingly, the major site of hydroxylation in these three straight chain fatty acids appears to be eight (C_{14} and C_{16}) or nine (C_{12}) methylenes removed from the carboxyl terminus, suggesting a role for this moiety in determining substrate binding and hydroxylation site selectivity. The CYP119 catalyzed oxidation of **1a**−**4a** was investigated, and ***Z*-1a**, **2a**, and ***E*-3a** were oxidized, although extremely poorly. In the oxidation of ***Z*-1a**, only the product from ω-2 oxidation was detected, while **2a** gave 80% ω-2 oxidation with the remainder of the hydroxylation occurring at the ω-3 carbon. The observed changes in the regiochemistry of oxidation again probably reflect the weakening of the pseudoallylic C−H bonds α to the cyclopropyl. ***E*-3a** gave a trace of a product, tentatively identified by MS fragmentation as bearing the hydroxyl at the ω-6 position. The levels of coupling were so low, however, that accurate quantitation was not possible, and the presence of other minor products cannot be excluded at this stage. The regiochemistry of oxidation of ***Z*-1a** is promising with respect to its use as a radical clock for this enzyme. The improvements in catalytic efficiency expected for the thermophilic CYP119 at elevated temperatures along with the recently reported thermally stable redox partners may allow ***Z*-1a** to be used as a mechanistic probe for this enzyme.[21,22,46]

Conclusion. The cyclopropyl containing fatty acids **1a**−**4a** reported here will hopefully prove useful as general mechanistic probes for the P450s and other enzymes. Their ability to distinguish between cationic and radical intermediates should prove particularly valuable in this regard. Here, they have demonstrated that a radical but not a cationic intermediate is formed during their oxidation catalyzed by $P450_{BM3}$ and $P450_{BioI}$. The rebound rate calculated from these data for the recombination of the intermediate carbon radical and the iron tethered oxygen was remarkably constant across both enzymes, probe structure, and position of oxidation. This suggests a homogeneity of mechanism across these enzymes with fatty acid substrates and perhaps in general across P450s with their natural substrates. Studies of **1a**−**4a** with P450s that are not normally involved in fatty acid metabolism and of analogues of them that are not usually good substrates for $P450_{BM3}$ and $P450_{BioI}$ may indicate how important the enzyme/substrate pair is to the observed mechanism. Our preliminary results with CYP119 also indicate that ***Z*-1a** may be of particular utility, if the level of oxidation can be increased sufficiently to allow investigation of any differential effect of temperature on the spin states of the iron oxo proposed to be important by two-state reactivity. These investigations are currently ongoing.

Experimental Procedures

General Methods and Instrumentation. See Supporting Information.

Enzyme Expression and Purification. $P450_{BioI}$,[19] $P450_{BM3}$[5], and CYP119[47] were expressed and purified as previously reported.

Enzymic Turnover. Incubation of cyclopropyl probes **1a**−**4a** with $P450_{BioI}$[18] (15 μM enzyme/1000 μM substrate/1250 μM NADPH) and $P450_{BM3}$[24] (2 μM enzyme/500 μM substrate/600 μM NADPH) was performed at 37 °C for 30 min as reported previously. Coupling is reported as the ratio of product formation to NADPH consumption (55−80% $P450_{BM3}$; 0−13% $P450_{BioI}$), and high percentage coupling corresponds to only small quantities of residual substrate remaining. Enzymatic turnover with CYP119: turnovers of **1a**−**4a** were performed in triplicate with two blanks using the following protocol: CYP119 (2 μM), fatty acid (500 μM), putidaredoxin reductase (2 μM), putidaredoxin (100 μM), catalase (1 μM), and superoxide dismutase (10 units) were combined in a Wheaton reaction vial. Aliquots of NADH (10 mM) were added to the solution every 2 h, while the turnover was incubated at 37 °C for at total of 6 h. Phenylacetic acid (final concentration 50 μM) was then added as an internal standard. The reaction was then acidified with HCl (1 M) extracted with diethyl ether (2 × 1 mL) and esterified with diazomethane. The turnover mixtures were concentrated before being analyzed by GC/MS. Blank turnovers were performed under identical conditions with the exception of either NADH or CYP119.

Analysis of Turnovers by GC/MS. See Supporting Information.

General Reaction Procedures. Procedure A: Addition of Deprotonated Terminal Alkyne. To a solution of terminal alkyne in THF (0.10 M) being stirred at −40 °C under a nitrogen atmosphere was added dropwise *n*-butyllithium (1.4 mol equiv). Following deprotonation of the alkyne after being stirred for 2 h, either HMPA (4% v/v) was added followed after 15 min by a solution of alkyl bromide in THF (3 mol equiv, 0.5 M) or a solution of aldehyde in THF (3 mol equiv, 0.5 M) was added directly without HMPA. The solution was warmed to room temperature and stirred for 12 h after which cold saturated, aqueous NH_4Cl (80 mL) was added to quench the reaction. The THF was removed in vacuo, and the aqueous layer was extracted with diethyl ether (3 × 50 mL). The organic layers were washed with 3 M aqueous LiCl (3 × 50 mL) and brine (50 mL) and dried over $MgSO_4$, and the solvent was removed in vacuo to afford the crude product.

Procedure B. Acidic Cleavage of THP-Ether. To a solution of THP-protected alcohol in methanol (∼0.1 M) was added *p*-TsOH (catalytic), and the reaction stirred for 2 h before being quenched by addition of solid $NaHCO_3$ (excess). The solvent was removed in vacuo, and the residue was partitioned between water (30 mL)/diethyl ether (30 mL). Following separation of the layers, the aqueous layer was extracted with diethyl ether (3 × 30 mL), and the combined organic layers were washed with brine (30 mL) and dried over $MgSO_4$, and the solvent was removed in vacuo to afford the desired alcohol.

Procedure C. Hydrogenation of Double/Triple Bonds. To a solution of alkene/alkyne in hexanes (∼0.3 M) and relevant hydrogenation catalyst (catalytic) was added the following: for reduction to single bonds, the catalyst used was either 5% Pd/C or PtO, while for reduction of alkynes to *Z*-alkenes, Lindlar's catalyst was used. The solution was introduced to a 1 atm hydrogen atmosphere and left to stir for 2 h at room temperature. The solution was filtered to remove the catalyst before being concentrated in vacuo to afford the desired alkane/alkene product.

Procedure D. Cyclopropanation of Alkene. To a solution of diiodomethane (6 mol equiv) in CH_2Cl_2 (∼1 M) being stirred at 0 °C under a nitrogen atmosphere was added diethyl zinc (3 mol equiv), and the reaction was stirred for 10 min. A

(45) Coupling is the ratio of product formation to NAD(P)H consumption.

(46) Puchkaev, A. V.; Ortiz de Montellano, P. R. *Arch. Biochem. Biophys.* **2005**, *434*, 169−177.

(47) Koo, L. S.; Tschirret-Guth, R. A.; Straub, W. E.; Moenne-Loccoz, P.; Loehr, T. M.; Ortiz de Montellano, P. R. *J. Biol. Chem.* **2000**, *275*, 14112−14123.

solution of alkene in CH_2Cl_2 (~0.5 M, 1 mol equiv) was added, and the reaction was stirred for 10 min before addition of a solution of TFA in CH_2Cl_2 (~0.5 M, 1 mol equiv). The reaction was allowed to stir at 0 °C for 1 h before being quenched by addition of cold saturated, aqueous NH_4Cl (20 mL). Following separation of the layers, the aqueous phase was extracted with CH_2Cl_2 (3 × 30 mL), and the combined organic layers were washed with brine (50 mL) before being dried over $MgSO_4$. Removal of the solvent in vacuo afforded the crude product.

Procedure E. Jones Oxidation. To a solution of alcohol in acetone (~ 0.1 M) being stirred at 0 °C was added Jones reagent (8 N) until the solution remained orange. The reaction was left to stir for 10 min before water (40 mL) was added, and the aqueous layer was extracted with hexanes (4 × 30 mL). The combined organic layers were washed with brine (50 mL), dried over $MgSO_4$, and concentrated in vacuo to afford the crude product.

Procedure F. Stabilized Wittig Reaction. To a solution of aldehyde in chloroform (~0.1 M) being stirred at room temperature was added methoxycarbonylmethylene(triphenyl)-phosphorane (2 mol equiv). The mixture was brought to reflux for 2 h before being cooled to room temperature. Following removal of the solvent in vacuo, the residue was passed through a silica gel plug to afford the crude product.

Procedure G. Base-Catalyzed Ester Cleavage. The ester was dissolved in methanol (~0.1 M), and aqueous 1 M LiOH (50% v/v) added dropwise while being stirred at room temperature. The reaction was left to stir for 12 h before being cooled to 0 °C and acidified to pH 1 with aqueous 1 M HCl. The aqueous phase was extracted with ethyl acetate (4 × 20 mL), and the combined organic layers were washed with brine (20 mL). After being dried over $MgSO_4$, the solvent was removed in vacuo to afford crude product.

Procedure H. PCC Oxidation of Alcohol. To a solution of alcohol in CH_2Cl_2 (0.1−0.01 M) being stirred at room temperature under a nitrogen atmosphere was added PCC (2 mol equiv). After 1 h, the reaction mixture was reduced in volume and passed through a silica gel plug to afford the crude aldehyde/ketone.

Procedure I. $NaBH_4$ Reduction of Ketone. To a solution of ketone in methanol (~0.1 M) was added either cerium trichloride hexahydrate (1.2 mol equiv) and $NaBH_4$ (5 mol equiv) over 10 min for conjugated ketones or $NaBH_4$ (5 mol equiv) over 10 min for nonconjugated ketones. The solution was allowed to stir for 2 h after which CH_2Cl_2 (20 mL) was added. The solution was then washed with aqueous, 5% oxalic acid solution (20 mL), and the layers were separated. The aqueous phase was extracted with CH_2Cl_2 (2 × 20 mL), and the combined organic layers were washed with saturated, aqueous $NaHCO_3$ solution (30 mL) and brine (30 mL), and the organic layer was dried over $MgSO_4$. Concentration in vacuo afforded the crude alcohol.

Procedure J. Grignard Addition to Carbonyl. To a solution of aldehyde/ketone in THF (0.2 M) being stirred at −40 °C under a nitrogen atmosphere was added dropwise a solution of the Grignard reagent (2.0 mol equiv) over 30 min. After being stirred overnight at room temperature, the solution was quenched with saturated, aqueous NH_4Cl (80 mL), and the layers were separated. The aqueous layer was extracted with diethyl ether (2 × 60 mL), and the combined organic layers were washed with brine (60 mL) and dried over $MgSO_4$, and the solvent was removed in vacuo to afford the crude product.

Procedure K. Ozonolysis of Double Bonds. Into a solution of alkene in CH_2Cl_2 (0.1 M) being stirred at −78 °C was bubbled excess ozone gas. After the ozonide had formed, the reaction was purged with O_2 and nitrogen before dimethyl sulfide (10% v/v) was added, and the reaction was warmed to room temperature. After being stirred at room temperature for 30 min, the solvent was removed in vacuo to afford the desired crude aldehyde.

Procedure L. Nonstabilized Wittig Reaction. To a solution of Wittig salt in diethyl ether (1.5 mol equiv, 0.1 M) being stirred at room temperature under nitrogen was added *n*-butyllithium (1.5 mol equiv). After 30 min, the reaction was cooled to −78 °C before a solution of aldehyde was added in diethyl ether (1.0 mol equiv, 0.25 M). After 2 h at −78 °C, the reaction was warmed to room temperature and quenched with ethanol (5 mL). Saturated, aqueous NH_4Cl (50 mL) and diethyl ether (50 mL) were added, and the layers were separated. The organic layer was extracted with diethyl ether (2 × 40 mL), washed with brine (40 mL), and dried over Na_2SO_4, and the solvent was removed in vacuo to afford the desired crude product.

1-(Tetrahydropyran-2′-yloxy)-tetradec-10-yne (19). Procedure A, 81% yield from 1-(tetrahydropyran-2′-yloxy)-undec-10-yne **18**.[48] ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.92 (3H, t, J_1 = 7.3 Hz), 1.10−1.80 (22H, m), 2.09 (4H, m), 3.20−3.90 (4H, 2m), 4.51 (1H, t, J_1 = 2.0 Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 13.4, 18.6, 19.6, 20.7, 22.5, 25.4, 26.1, 28.8, 29.0, 29.1, 29.36, 29.40, 29.7, 30.7, 62.2, 67.6, 79.9, 80.3, 98.7. GC/MS: 294 ($M^{+\bullet}$, 0.1), 251 (1.1), 123 (2.3), 109 (4.2), 101 (22.5), 85 (100), 55 (33.2), 41 (57.4). Anal. Calcd. for $C_{19}H_{34}O_2$: C, 77.50; H, 11.64. Found: C, 77.73; H, 11.28.

Tetradec-10-(*Z*)-en-1-ol (*Z*-20). Procedures B and C, 86% yield from **19**, >98% *Z*. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.85 (3H, t, J_1 = 7.4 Hz), 1.20−1.40 (13H, m), 1.45 (2H, quint, J_1 = 4.7 Hz), 1.53 (2H, quint, J_1 = 7.1 Hz), 2.09 (4H, m), 3.60 (2H, d of d, J_1 = 6.5 Hz, J_2 = 5.4 Hz), 5.36 (2H, m). ^{13}C NMR (100 MHz, $CDCl_3$) δ 13.6, 22.7, 25.7, 28.8, 29.3, 29.40, 29.44, 29.5, 29.7, 32.8, 34.7, 63.0, 130.1, 130.5. GC/MS: 166 (1.5), 138 (1.8), 123 (2.4), 110 (4.8), 95 (12.8), 68 (45.1), 55 (91.1), 41 (100). Anal. Calcd. for $C_{14}H_{28}O$: C, 79.18; H, 13.29. Found: C, 79.01; H, 13.24.

Tetradec-10-(*E*)-en-1-ol (*E*-20). The alcohol from **19** (0.550 g, 1.87 mmol) was prepared employing the general THP-cleavage procedure B (0.337 g). To liquid ammonia (15 mL) being stirred at −78 °C was added a solution of crude alcohol (0.337 g) in *t*-butyl alcohol/THF (3:5, 8 mL). After 5 min, lithium metal (0.080 g) was added, and the reaction was left to stir for 1.5 h. The ammonia was then allowed to evaporate after which saturated, aqueous NH_4Cl (30 mL) was added to quench the reaction. The aqueous layer was extracted with hexanes (4 × 30 mL) that were in turn washed with brine (3 × 30 mL). The organic layer was dried over $MgSO_4$ and concentrated in vacuo and purified by flash chromatography (silica gel, 20% ethyl acetate in hexanes) to afford ***E*-20** (0.269 g, 1.27 mmol, 68% yield, >95% *Z*) as a clear, colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.87 (3H, t, J_1 = 7.4 Hz), 1.20−1.40 (13H, m), 1.47 (2H, quint, J_1 = 5.4 Hz), 1.53 (2H, quint, J_1 = 7.2 Hz), 1.97 (4H, quint, J_1 = 6.5 Hz), 3.61 (2H, d of d, J_1 = 6.6 Hz, J_2 = 5.4 Hz), 5.33 (2H, m). ^{13}C NMR (100 MHz, $CDCl_3$) δ 13.8, 22.9, 25.7, 27.2, 29.25, 29.29, 29.39, 29.44, 29.5, 29.7, 32.8, 63.0, 129.6, 130.0. GC/MS: 166 (1.5), 138 (1.5), 123 (2.2), 110 (4.1), 95 (12.5), 68 (45.1), 55 (81.3), 41 (100). Anal. Calcd. for $C_{14}H_{28}O$: C, 79.18; H, 13.29. Found: C, 78.78; H, 12.91.

9-(*Z*-2-Propylcyclopropyl)-nonan-1-ol (*Z*-21). Procedure D, 97% yield from ***Z*-20**. ^{1}H NMR (200 MHz, $CDCl_3$) δ −0.42 (1H, d of d of d, J_1 = 8.5 Hz, J_2 = 8.0 Hz, J_3 = 4.0 Hz), 0.54 (3H, m), 0.84 (3H, t, J_1 = 7.0 Hz), 1.00−1.35 (18H, m), 1.42 (2H, quint, J_1 = 7.6 Hz), 2.87 (1H, br s), 3.51 (2H, t, J_1 = 6.5 Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 10.7, 14.0, 15.3, 15.5, 23.1, 25.7, 28.6, 29.3, 29.4, 29.48, 29.53, 30.1, 30.7, 32.6, 62.5. GC/MS: 208 (0.2), 183 (0.5), 123 (2.9), 109 (7.3), 95 (18.6), 82 (33.4), 41 (100). Anal. Calcd. for $C_{15}H_{30}O$: C, 79.58; H, 13.36. Found: C, 79.29; H, 13.17.

9-(*E*-2-Propylcyclopropyl)-nonan-1-ol (*E*-21). Procedure D, 100% yield from ***E*-20**. ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.03 (2H, t, J_1 = 6.5 Hz), 0.32 (2H, m), 0.83 (3H, t, J_1 = 7.2 Hz), 1.00−1.35 (18H, m), 1.44 (2H, quint, J_1 = 6.5 Hz), 2.88 (1H,

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br s), 3.52 (2H, t, $J_1 = 6.5$ Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 11.6, 13.9, 18.4, 18.6, 22.6, 25.7, 29.38, 29.40, 29.42, 29.56, 29.59, 32.6, 34.2, 36.4, 62.5. GC/MS: 208 (0.2), 183 (0.7), 123 (2.8), 109 (7.0), 95 (17.1), 81 (30.9), 41 (100). Anal. Calcd. for $C_{15}H_{30}O$: C, 79.58; H, 13.36. Found: C, 79.19; H, 13.48.

9-(*Z*-2-Propylcyclopropyl)-nonanoic Acid (*Z*-1a). Procedure E, 70% yield from ***Z*-21**, mp 40.0−42.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.35 (1H, d of d of d, $J_1 = 8.5$ Hz, $J_2 = 8.0$ Hz, $J_3 = 4.0$ Hz), 0.55 (1H, d of d of d, $J_1 = 5.2$ Hz, $J_2 = 5.2$ Hz, $J_3 = 4.0$ Hz), 0.64 (2H, m), 0.90 (3H, t, $J_1 = 7.6$ Hz), 1.10−1.40 (16H, m), 1.61 (2H, quint, $J_1 = 6.4$ Hz), 2.32 (2H, t, $J_1 = 7.6$ Hz), 10.0 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 10.9, 14.1, 15.5, 15.7, 23.3, 24.7, 28.7, 29.0, 29.2, 29.5, 29.6, 30.1, 30.9, 34.1, 180.6. Anal. Calcd. for $C_{15}H_{28}O_2$: C, 74.95; H, 11.74. Found: C, 75.16; H, 12.11.

Methyl 9-(*Z*-2-Propylcyclopropyl)-nonanoate (*Z*-1). Prepared by derivatization of ***Z*-1a** with an ethereal solution of diazomethane. GC/MS: 254 ($M^{+\bullet}$, 0.2), 222 (2.5), 180 (2.9), 152 (2.6), 138 (3.8), 123 (5.8), 110 (7.6), 41 (100).

9-(*E*-2-Propylcyclopropyl)-nonanoic Acid (*E*-1a). Procedure E, 91% yield from ***E*-21**, mp 40.0−42.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.11 (2H, d of d, $J_1 = 6.5$ Hz, $J_2 = 6.5$ Hz), 0.34 (2H, d of d of d of d, $J_1 = 17.2$ Hz, $J_2 = 8.9$ Hz, $J_3 = 6.3$ Hz, $J_4 = 4.5$ Hz), 0.89 (3H, t, $J_1 = 7.6$ Hz), 1.10−1.40 (16H, m), 1.61 (2H, quint, $J_1 = 7.2$ Hz), 2.32 (2H, t, $J_1 = 7.5$ Hz), 10.0 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 11.7, 14.0, 18.5, 18.7, 22.8, 24.6, 29.0, 29.2, 29.4, 29.5, 29.6, 34.1, 34.3, 36.5, 180.7. Anal. Calcd. for $C_{15}H_{28}O_2$: C, 74.95; H, 11.74. Found: C, 74.90; H, 12.10.

Methyl 9-(*E*-2-Propylcyclopropyl)-nonanoate (*E*-1). Prepared by derivatization of ***E*-1a** with an ethereal solution of diazomethane. GC/MS: 254 ($M^{+\bullet}$, 0.2), 222 (2.6), 180 (2.9), 152 (2.4), 138 (3.7), 123 (5.4), 110 (7.2), 41 (100).

11-(*Z*-2-Propylcyclopropyl)-undecanoic Acid (*Z*-3a). To a solution of oxalyl chloride (0.105 mL, 1.11 mmol) in CH_2Cl_2 (5 mL) being stirred at −78 °C under a nitrogen atmosphere was added dropwise DMSO (0.205 mL, 2.76 mmol). After being stirred for 10 min, a solution of ***Z*-21** (0.140 g, 0.614 mmol) in CH_2Cl_2 (1 mL) was added dropwise, and the reaction was left to stir at −78 °C for 1 h. Triethylamine (1.33 mL, 9.82 mmol) was added, and the reaction was allowed to warm to room temperature. Brine (50 mL) and diethyl ether (20 mL) were added, and following separation of the layers, the aqueous phase was extracted with diethyl ether (2 × 20 mL). The combined organic layers were dried over $MgSO_4$, and the solvent was removed in vacuo to yield aldehyde (0.102 g) that was used without further purification. Chain extension of the aldehyde (0.102 g) was achieved via Wittig condensation employing general procedure F, and the product was then hydrogenated using the general procedure C using 5% Pd/C catalyst. Base cleavage of the ester was accomplished using the general procedure G. The residue was purified using flash chromatography (silica gel, 20% ethyl acetate in hexanes, 1% acetic acid) to afford ***Z*-3a** (0.030 g, 0.111 mmol, 10% yield over four steps) as a white solid, mp 45.0−47.0 °C). ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.34 (1H, d of d of d, $J_1 = 8.5$ Hz, $J_2 = 8.0$ Hz, $J_3 = 4.0$ Hz), 0.54 (1H, d of d of d, $J_1 = 5.2$ Hz, $J_2 = 5.2$ Hz, $J_3 = 4.0$ Hz), 0.64 (2H, m), 0.90 (3H, t, $J_1 = 7.0$ Hz), 1.10−1.40 (20H, m), 1.62 (2H, quint, $J_1 = 6.4$ Hz), 2.32 (2H, t, $J_1 = 7.4$ Hz), 10.9 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 10.9, 14.1, 15.5, 15.7, 23.3, 24.7, 28.7, 29.0, 29.2, 29.4, 29.5, 29.58, 29.60, 30.1, 30.9, 34.1, 180.6. Anal. Calcd. for $C_{17}H_{32}O_2$: C, 76.06; H, 12.01. Found: C, 76.09; H, 12.30.

Methyl 11-(*Z*-2-Propylcyclopropyl)-undecanoate (*Z*-3). Prepared by derivatization of ***Z*-3a** with an ethereal solution of diazomethane. GC/MS: 282 ($M^{+\bullet}$, 0.1), 251 (2.5), 250 (4.1), 208 (2.2), 166 (3.0), 110 (6.9), 97 (16.5), 41 (100).

11-(*E*-2-Propylcyclopropyl)-undecanoic Acid (*E*-3a). Procedure identical to ***Z*-3a**, 15% yield over four steps, mp 45.0−47.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.12 (2H, d of d, $J_1 = 6.5$ Hz, $J_2 = 6.5$ Hz), 0.35 (2H, d of d of d of d, $J_1 = 17.2$ Hz, $J_2 = 8.9$ Hz, $J_3 = 6.3$ Hz, $J_4 = 4.5$ Hz), 0.88 (3H, t, $J_1 = 7.3$ Hz), 1.10−1.40 (20H, m), 1.60 (2H, quint, $J_1 = 7.1$ Hz), 2.32 (2H, t, $J_1 = 7.5$ Hz), 8.10 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 11.7, 14.0, 18.5, 18.7, 22.8, 24.7, 29.1, 29.2, 29.4, 29.5, 29.6, 29.66, 29.70, 34.0, 34.3, 36.6, 179.9. Anal. Calcd. for $C_{17}H_{32}O_2$: C, 76.06; H, 12.01. Found: C, 75.77; H, 11.99.

Methyl 11-(*E*-2-Propylcyclopropyl)-undecanoate (*E*-3). Prepared by derivatization of ***E*-3a** with an ethereal solution of diazomethane. GC/MS: 282 ($M^{+\bullet}$, 0.3), 251 (3.2), 250 (4.8), 208 (2.7), 166 (3.5), 110 (7.5), 97 (17.1), 55 (100).

12-Cyclopropyl-1-(tetrahydropyran-2′-yloxy)-dodec-2-yne (23). Procedure A, 85% yield from 1-bromo-9-cyclopropylnonane **22** (See Supporting Information). ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.05 (2H, d of d of d, $J_1 = 4.8$ Hz, $J_2 = 4.8$ Hz, $J_3 = 1.5$ Hz), 0.34 (2H, d of d of d, $J_1 = 8.4$ Hz, $J_2 = 8.4$ Hz, $J_3 = 1.9$ Hz), 0.61 (1H, m), 1.10−1.50 (22H, m), 2.18 (2H, t of t, $J_1 = 3.6$ Hz, $J_2 = 1.1$ Hz), 3.50 (1H, m), 3.81 (1H, m), 4.21 (2H, m), 4.78 (1H, t, $J_1 = 3.4$ Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 4.3 (2C), 10.9, 18.8, 19.1, 25.4, 28.6, 28.8, 29.1, 29.4, 29.48, 29.51, 29.6, 30.3, 34.7, 54.6, 61.9, 75.7, 86.7, 96.6. GC/MS: 306 ($M^{+\bullet}$, 0.1), 135 (2.6), 121 (4.2), 111 (7.8), 101 (19.7), 95 (23.3), 85 (100), 55 (52.8). Anal. Calcd. for $C_{20}H_{34}O_2$: C, 78.38; H, 11.18. Found: C, 78.59; H, 10.78.

12-Cyclopropyl-1-(tetrahydropyran-2′-yloxy)-dodecane (24). Procedure C, 99% yield from **23**. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.04 (2H, d of d of d, $J_1 = 4.8$ Hz, $J_2 = 4.8$ Hz, $J_3 = 1.5$ Hz), 0.34 (2H, d of d of d, $J_1 = 8.4$ Hz, $J_2 = 8.4$ Hz, $J_3 = 1.9$ Hz), 0.61 (1H, m), 1.16 (2H, q, $J_1 = 7.2$ Hz), 1.20−1.40 (26H, m), 3.25−3.95 (4H, m), 4.56 (1H, t, $J_1 = 3.5$ Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 4.3 (2C), 10.9, 18.9, 19.7, 25.5, 26.2, 26.7, 29.4, 29.49, 29.54, 29.6, 29.7, 29.8, 30.8, 34.8, 53.9, 61.9, 67.7, 98.8. GC/MS: 310 ($M^{+\bullet}$, 0.3), 153 (1.0), 123 (1.2), 101 (10.4), 85 (100), 55 (36.9), 43 (32.4), 41 (50.5). Anal. Calcd. for $C_{20}H_{38}O_2$: C, 77.36; H, 12.33. Found: C, 77.08; H, 12.07.

12-Cyclopropyldodecan-1-ol (25). Procedure B, 98% yield from **24**. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.04 (2H, d of d of d, $J_1 = 4.8$ Hz, $J_2 = 4.8$ Hz, $J_3 = 1.5$ Hz), 0.34 (2H, d of d of d, $J_1 = 8.4$ Hz, $J_2 = 8.4$ Hz, $J_3 = 1.9$ Hz), 0.61 (1H, m), 1.16 (2H, q, $J_1 = 7.2$ Hz), 1.20−1.40 (21H, m), 3.60 (2H, t, $J_1 = 6.8$ Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 4.3 (2C), 10.9, 18.9, 25.5, 26.2, 26.7, 29.4, 29.5, 29.6, 29.7, 29.8, 30.8, 34.8, 62.5. GC/MS: 180 (0.1), 152 (1.0), 124 (3.6), 123 (3.4), 109 (7.0), 95 (16.1), 55 (78.6), 41 (100). Anal. Calcd. for $C_{15}H_{30}O$: C, 79.58; H, 13.36. Found: C, 79.70; H, 13.49.

12-Cyclopropyldodecanoic Acid (2a). Procedure E, 89% yield from **25**, mp 48.0−49.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.04 (2H, d of d of d, $J_1 = 4.8$ Hz, $J_2 = 4.8$ Hz, $J_3 = 1.5$ Hz), 0.35 (2H, d of d of d, $J_1 = 8.4$ Hz, $J_2 = 8.4$ Hz, $J_3 = 1.9$ Hz), 0.61 (1H, m), 1.15 (2H, q, $J_1 = 7.2$ Hz), 1.20−1.55 (16H, m), 1.61 (2H, quint, $J_1 = 7.6$ Hz), 2.33 (2H, t, $J_1 = 7.4$ Hz), 10.10 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 4.3 (2C), 10.9, 24.7, 29.1, 29.2, 29.4, 29.5, 29.58, 29.63, 29.65, 29.67, 34.1, 37.8, 180.5. Anal. Calcd. for $C_{15}H_{28}O_2$: C, 74.95; H, 11.74. Found: C, 75.00; H, 12.10.

Methyl 12-Cyclopropyldodecanoate (2). Prepared by derivatization of **2a** with an ethereal solution of diazomethane. GC/MS: 224 (0.4), 180 (2.9), 152 (2.9), 138 (4.2), 123 (6.3), 110 (8.9), 98 (13.7), 41 (100).

Methyl 14-Cyclopropyltetradec-2-enoate (26). Procedures H and F, 57% yield from **25**. ^{1}H NMR (200 MHz, $CDCl_3$) δ −0.04 (2H, d of d of d, $J_1 = 4.8$ Hz, $J_2 = 4.8$ Hz, $J_3 = 1.5$ Hz), 0.36 (2H, d of d of d, $J_1 = 8.4$ Hz, $J_2 = 8.4$ Hz, $J_3 = 1.9$ Hz), 0.62 (1H, m), 1.17 (2H, q, $J_1 = 7.2$ Hz), 1.20−1.45 (16H, m), 1.61 (2H, quint, $J_1 = 7.6$ Hz), 2.15 (2H, d of q, $J_1 = 1.5$ Hz, $J_2 = 7.0$ Hz), 3.68 (3H, s), 5.78 (1H, d of t, $J_1 = 15.6$ Hz, $J_2 = 1.5$ Hz), 6.93 (1H, d of t, $J_1 = 15.8$ Hz, $J_2 = 7.0$ Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 4.3 (2C), 10.9, 24.7, 27.9, 29.1, 29.3, 29.4, 29.55, 29.58, 29.64, 29.7, 32.2, 37.2, 51.4, 120.7, 149.9, 167.2. Anal. Calcd. for $C_{18}H_{32}O_2$: C, 77.09; H, 11.50. Found: C, 77.01; H, 11.23.

14-Cyclopropyltetradecanoic Acid (4a). Procedures C and G, 64% yield from **26**, mp 54.0−55.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.04 (2H, d of d of d, $J_1 = 4.8$ Hz, $J_2 = 4.8$

Hz, J_3 = 1.5 Hz), 0.36 (2H, d of d of d, J_1 = 8.4 Hz, J_2 = 8.4 Hz, J_3 = 1.9 Hz), 0.62 (1H, m), 1.17 (2H, q, J_1 = 7.2 Hz), 1.20−1.45 (20H, m), 1.61 (2H, quint, J_1 = 7.6 Hz), 2.32 (2H, t, J_1 = 7.6 Hz), 8.80 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 4.3 (2C), 10.9, 24.7, 29.1, 29.2, 29.4, 29.55, 29.58, 29.63, 29.65, 29.66, 29.68, 29.71, 34.1, 37.8, 180.1. Anal. Calcd. for $C_{17}H_{32}O_2$: C, 76.06; H, 12.01. Found: C, 75.96; H, 11.63.

Methyl 14-Cyclopropyltetradecanoate (4). Prepared by derivatization of **4a** with an ethereal solution of diazomethane. GC/MS: 282 ($M^{+\bullet}$, 0.1), 250 (2.9), 208 (2.7), 166 (3.2), 152 (3.2), 111 (7.3), 98 (14.3), 41 (100).

1-(Tetrahydropyran-2′-yloxy)-tetradec-10-yn-12-ol (27). Procedure A, 99% yield from **18**.[48] ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.90 (3H, t, J_1 = 6.9 Hz), 0.20−1.70 (21H, m), 1.82 (2H, m), 2.17 (2H, t of d, J_1 = 7.1 Hz, J_2 = 2.0 Hz), 3.30−3.90 (4H, m), 4.27 (1H, t of t, J_1 = 6.4 Hz, J_2 = 1.9 Hz), 4.55 (1H, t, J_1 = 2.6 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 9.4, 18.6, 19.7, 25.5, 26.2, 28.6, 29.0, 29.37, 29.39, 29.7, 29.8, 30.8, 31.2, 62.3, 63.9, 67.6, 81.1, 85.6, 98.8. GC/MS: 263 (0.1), 251 (0.3), 197 (5.6), 135 (1.8), 109 (3.6), 101 (23.3), 85 (100), 41 (47.3). Anal. Calcd. for $C_{19}H_{34}O_3$: C, 73.50; H, 11.04. Found: C, 73.77; H, 11.38.

1-(Tetrahydropyran-2′-yloxy)-tetradec-10-(*Z*)-en-12-ol (*Z*-28). Procedure C, 100% yield from **27**, >98% *Z*. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.86 (3H, t, J_1 = 7.6 Hz), 1.20−1.70 (23H, m), 2.03 (2H, quint, J_1 = 6.9 Hz), 3.30−3.85 (4H, m), 4.30 (1H, t of d, J_1 = 9.6 Hz, J_2 = 7.4 Hz), 4.53 (1H, t, J_1 = 2.6 Hz), 5.31 (1H, t of d, J_1 = 9.0 Hz, J_2 = 7.6 Hz), 5.44 (1H, d of t, J_1 = 11.0 Hz, J_2 = 7.4 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 9.6, 19.6, 25.4, 26.2, 27.7, 29.2, 29.4, 29.5, 29.6, 29.7, 30.1, 30.3, 30.7, 62.2, 67.6, 69.0, 98.7, 132.3, 132.4. GC/MS: 201 (0.1), 199 (0.3), 135 (0.6), 123 (1.1), 109 (3.3), 101 (20.9), 85 (100), 41 (58.2). Anal. Calcd. for $C_{19}H_{36}O_3$: C, 73.03; H, 11.61. Found: C, 72.95; H, 11.73.

1-(Tetrahydropyran-2′-yloxy)-tetradec-10-(*E*)-en-12-ol (*E*-28). To a solution of lithium aluminum hydride (0.090 g, 24.6 mmol) in THF (60 mL) being stirred at 0 °C under a nitrogen atmosphere was added dropwise a solution of **27** (0.765 g, 2.46 mmol) in THF (20 mL). The solution was brought to reflux for 1 h before the solution was cooled to room temperature, and sodium sulfate decahydrate (excess) was added over 2 h to quench the reaction. The solution was then filtered, and the salts were washed with ethyl acetate (5 × 30 mL). Following concentration in vacuo, the residue was purified by flash chromatography (silica gel, 10% ethyl acetate in hexanes) to afford ***E*-28** (0.760 g, 2.43 mmol, 99% yield, >99% *E*) as a clear, colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.87 (3H, t, J_1 = 6.9 Hz), 1.20−1.70 (23H, m), 2.00 (2H, q, J_1 = 6.8 Hz), 3.30−3.85 (5H, m), 4.54 (1H, t, J_1 = 2.0 Hz), 5.41 (1H, d of d of t, J_1 = 15.4 Hz, J_2 = 7.1 Hz, J_3 = 1.1), 5.60 (1H, t of t of d, J_1 = 15.4 Hz, J_2 = 6.7 Hz, J_3 = 0.9 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 9.8, 14.0, 19.7, 25.5, 26.2, 27.8, 29.1, 29.4, 29.5, 29.7, 30.1, 30.8, 36.6, 62.3, 67.7, 74.5, 98.8, 132.4, 132.7. GC/MS: 283 (0.1), 199 (0.7), 135 (0.9), 123 (1.5), 109 (5.0), 101 (20.5), 85 (100), 41 (48.3). Anal. Calcd. for $C_{19}H_{36}O_3$: C, 73.03; H, 11.61. Found: C, 72.98; H, 11.61.

9-(2-(1-Hydroxypropyl)-*Z*-cyclopropyl)-1-(tetrahydropyran-2′-yloxy)-nonane (*Z*-29). Procedure D, 100% yield from ***Z*-28**. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.01 (1H, d of d, J_1 = 5.2 Hz, J_2 = 5.2 Hz), 0.66 (1H, t of d of d, J_1 = 8.4 Hz, J_2 = 4.4 Hz, J_3 = 0.4 Hz), 0.75−0.85 (2H, m), 0.95 (3H, t, J_1 = 7.6 Hz), 1.20−1.80 (25H, m), 3.08 (1H, d of t, J_1 = 8.1 Hz, J_2 = 6.2 Hz), 3.30−3.85 (4H, m), 4.53 (1H, t, J_1 = 2.8 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 9.4, 9.9, 16.8, 19.6, 22.8, 25.4, 26.2, 29.1, 29.40, 29.43, 29.49, 29.50, 29.7, 30.1, 30.7, 36.6, 62.2, 67.6, 73.7, 98.7. GC/MS: 213 (7.2), 121 (1.2), 109 (2.6), 101 (14.1), 95 (7.4), 85 (100), 57 (30.0), 41 (51.6). Anal. Calcd. for $C_{20}H_{38}O_3$: C, 73.57; H, 11.73. Found: C, 73.23; H, 11.95.

9-(2-(1-Hydroxypropyl)-*E*-cyclopropyl)-1-(tetrahydropyran-2′-yloxy)-nonane (*E*-29). Procedure D, 98% yield from ***E*-28**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.23 (1H, m), 0.38 (1H, m), 0.58 (2H, m), 0.95 (3H, t, J_1 = 7.4 Hz, 1.15−1.80 (25H, m), 2.79 (1H, q of d, J_1 = 6.9 Hz, J_2 = 1.2 Hz), 3.30−3.90 (4H, m), 4.54 (1H, t, J_1 = 1.7 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 9.7, 10.0, 17.0, 19.7, 25.1, 25.4, 26.2, 29.40, 29.43, 29.5, 29.7, 29.8, 30.0, 30.1, 30.8, 36.6, 62.3, 67.6, 77.7, 98.8. GC/MS: 241 (0.5), 213 (9.4), 109 (2.9), 101 (11.3), 95 (7.4), 85 (100), 57 (33.5), 41 (51.1). Anal. Calcd. for $C_{20}H_{38}O_3$: C, 73.57; H, 11.73. Found: C, 73.40; H, 11.94.

***anti*-9-(2-(1-Hydroxypropyl)-*Z*-cyclopropyl)-nonanoic Acid (*Z*-5a).** Procedures B, E, and I, 83% yield from ***Z*-29**, mp 64.0−65.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.10 (1H, d of d, J_1 = 5.2 Hz, J_2 = 5.2 Hz), 0.68 (1H, d of d of d, J_1 = 8.4 Hz, J_2 = 8.4 Hz, J_3 = 4.5 Hz), 0.77 (1H, d of d of d of d, J_1 = 11.6 Hz, J_2 = 5.5 Hz, J_3 = 5.0 Hz, J_4 = 3.3 Hz), 0.86 (1H, d of d of d, J_1 = 9.1 Hz, J_2 = 4.5 Hz, J_3 = 1.0 Hz), 0.95 (3H, t, J_1 = 7.5 Hz), 1.20−1.40 (13H, m), 1.61 (4H, m), 2.30 (2H, t, J_1 = 7.6 Hz), 3.11 (1H, d of t, J_1 = 8.0 Hz, J_2 = 6.6 Hz), 5.56 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 9.7, 10.1, 22.4, 24.7, 28.6, 29.0, 29.2, 29.3, 29.36, 29.41, 29.5, 30.1, 34.1, 74.5, 179.4. Anal. Calcd. for $C_{15}H_{28}O_3$: C, 70.27; H, 11.01. Found: C, 70.01; H, 10.77.

Methyl *anti*-9-(2-(1-Hydroxypropyl)-*Z*-cyclopropyl)-nonanoate (*Z*-5). Prepared by derivatization of ***Z*-5a** with an ethereal solution of diazomethane. GC/ MS: 252 (0.4), 241 (4.5), 209 (8.1), 173 (9.5), 169 (24.0), 149 (11.1), 81 (48.9), 41 (100).

***anti*-9-(2-(1-Hydroxypropyl)-*E*-cyclopropyl)-nonanoic Acid (*E*-5a).** Procedures B, E, and I, 69% yield from ***E*-29**, mp 64.0−65.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.30 (2H, d of d of t, J_1 = 25.3 Hz, J_2 = 7.1 Hz, J_3 = 5.8 Hz), 0.60 (2H, m), 0.93 (3H, t, J_1 = 7.5 Hz), 1.19 (2H, q of d, J_1 = 6.8 Hz, J_2 = 2.6 Hz), 1.22−1.40 (11H, m), 1.57 (2H, quint, J_1 = 7.2 Hz), 1.60 (2H, quint of d, J_1 = 5.7 Hz, J_2 = 2.0 Hz), 2.30 (2H, t, J_1 = 7.5 Hz), 2.82 (1H, q of d, J_1 = 5.6 Hz, J_2 = 1.2 Hz), 5.88 (1H, br s). ^{13}C NMR (100 MHz, $CDCl_3$) δ 10.0, 16.5, 17.1, 24.7, 29.1, 29.2, 29.3, 29.4, 29.6, 29.8, 30.0, 33.7, 34.1, 77.9, 179.4. Anal. Calcd. for $C_{15}H_{28}O_3$: C, 70.27; H, 11.01. Found: C, 70.22; H, 11.36.

Methyl *anti*-9-(2-(1-Hydroxypropyl)-*E*-cyclopropyl)-nonanoate (*E*-5a). Prepared by derivatization of ***E*-5a** with an ethereal solution of diazomethane. GC/MS: 269 (0.4), 252 (0.3), 241 (10.4), 209 (16.4), 173 (10.8), 169 (11.7), 149 (9.0), 41 (100).

***anti*-11-(2-(1-Hydroxypropyl)-*Z*-cyclopropyl)-undecanoic Acid (*Z*-7a).** Procedures B, H, F, C, I, and G, 19% yield over six steps from ***Z*-29**, mp 68.0−69.0 °C. ^{1}H NMR (200 MHz, $CDCl_3$) δ −0.09 (1H, d of d, J_1 = 5.2 Hz, J_2 = 5.2 Hz), 0.60−0.90 (3H, m), 0.96 (3H, t, J_1 = 7.4 Hz), 1.20−1.40 (16H, m), 1.60 (4H, m), 2.07 (1H, br s), 2.32 (2H, t, J_1 = 7.4 Hz), 3.13 (1H, d of t, J_1 = 9.0 Hz, J_2 = 6.6 Hz), 5.95 (1H, br s). ^{13}C NMR (50 MHz, $CDCl_3$) δ 9.7, 10.1, 15.5, 22.4, 24.6, 28.6, 28.9, 29.1, 29.3, 29.4, 29.46, 29.54, 30.1, 30.6, 34.0, 74.5, 179.6. Anal. Calcd. for $C_{17}H_{32}O_3$: C, 71.79; H, 11.34. Found: C, 72.07; H, 11.11.

Methyl *anti*-11-(2-(1-Hydroxypropyl)-*Z*-cyclopropyl)-undecanoate (*Z*-7). Prepared by derivatization of ***Z*-7a** with an ethereal solution of diazomethane. GC/MS: 280 (0.4), 269 (4.0), 237 (9.2), 201 (3.9), 197 (8.3), 177 (5.2), 121 (9.0), 41 (100).

***anti*-11-(2-(1-Hydroxypropyl)-*E*-cyclopropyl)-undecanoic Acid (*E*-7a).** Procedures B, H, F, C, I, and G, 17% yield over six steps from ***E*-29**, mp 68.0−69.0 °C. ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.32 (2H, d of d of t, J_1 = 25.3 Hz, J_2 = 7.1 Hz, J_3 = 5.8 Hz), 0.59 (2H, m), 0.94 (3H, t, J_1 = 7.5 Hz), 1.10−1.40 (17H, m), 1.59 (4H, m), 2.31 (2H, t, J_1 = 7.3 Hz), 2.82 (1H, q of d, J_1 = 6.6 Hz, J_2 = 1.2 Hz), 5.22 (1H, br s). ^{13}C NMR (50 MHz, $CDCl_3$) δ 10.0, 16.6, 24.6, 25.0, 29.0, 29.1, 29.25, 29.33, 29.4, 29.5, 29.6, 29.8, 30.0, 33.7, 33.9, 77.9, 179.3. Anal. Calcd. for $C_{17}H_{32}O_3$: C, 71.79; H, 11.34. Found: C, 71.69; H, 11.73.

Methyl *anti*-11-(2-(1-Hydroxypropyl)-*E*-cyclopropyl)-undecanoate (*E*-7). Prepared by derivatization of ***E*-7a** with an ethereal solution of diazomethane. GC/MS: 280 (0.3), 269 (7.8), 237 (16.8), 201 (4.2), 177 (3.9), 135 (7.8), 121 (8.6), 41 (100).

12,12′-Dimethoxydodecanal (30). Into a solution of cyclododecene (3.000 g, 18.07 mmol) in 20% methanol in CH_2-

Cl_2 (40 mL) being stirred at −40 °C was bubbled excess ozone gas. After the ozonide had formed, the reaction was purged with oxygen and nitrogen before *p*-TsOH (0.350 g, 10% w/w) was added, and the reaction was allowed to warm to room temperature. After being stirred at room temperature for 90 min, solid $NaHCO_3$ (2.000 g) was added followed 15 min later by dimethyl sulfide (4.00 mL), and the reaction was stirred at room temperature overnight under nitrogen. Brine (80 mL) was added, and the solution was extracted with CH_2Cl_2 (2 × 60 mL) following separation of the layers. The organic layers were washed with brine (80 mL) and dried over $MgSO_4$, and the solvent removed in vacuo to afford **30** (3.730 g, 16.59 mmol, 92% yield) as a clear, colorless oil. ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.10−1.30 (14H, m), 1.59 (4H, m), 2.35 (2H, t of d, J_1 = 7.3 Hz, J_2 = 1.8 Hz), 3.23 (6H, s), 4.28 (1H, t, J_1 = 5.6 Hz), 9.68 (1H, t, J_1 = 1.8 Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 21.9, 24.4, 29.0, 29.20, 29.24, 29.31, 29.32, 29.4, 32.3, 43.8, 52.4 (2C), 104.4, 202.7. GC/MS: 213 (2.9), 163 (0.8), 121 (1.4), 95 (5.0), 75 (100). Anal. Calcd. for $C_{14}H_{28}O_3$: C, 69.08; H, 11.21. Found: C, 68.81; H, 11.55.

14,14′-Dimethoxytetradec-1-en-3-ol (31). Procedure J, 92% yield from **30**. ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.10−1.40 (16H, m), 1.51 (4H, m), 1.80 (1H, br s), 3.26 (6H, s), 4.03 (1H, q, J_1 = 6.3 Hz), 4.31 (1H, t, J_1 = 5.6 Hz), 5.10 (2H, m), 5.82 (1H, m). ^{13}C NMR (50 MHz, $CDCl_3$) δ 24.5, 25.3, 29.3, 29.38, 29.45, 29.46, 29.48, 29.50, 32.4, 37.0, 52.5 (2C), 73.1, 104.5, 114.4, 141.3. GC/MS: 223 (0.8), 152 (1.0), 109 (4.2), 95 (8.0), 75 (100). Anal. Calcd. for $C_{16}H_{32}O_3$: C, 70.54; H, 11.84. Found: C, 70.51; H, 11.56.

1-Cyclopropyl-12,12′-dimethoxydodecan-1-ol (32). Procedure D, 92% yield from **31**. ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.21 (2H, d of d of d of d, J_1 = 22.0 Hz, J_2 = 13.1 Hz, J_3 = 4.9 Hz, J_4 = 4.1 Hz), 0.48 (2H, d of d of d of d, J_1 = 17.2 Hz, J_2 = 12.0 Hz, J_3 = 7.7 Hz, J_4 = 3.9 Hz), 0.87 (1H, d of d of d of d, J_1 = 16.3 Hz, J_2 = 8.4 Hz, J_3 = 5.1 Hz, J_4 = 5.1 Hz), 1.20−1.60 (21H, m), 2.82 (1H, d of t, J_1 = 8.4 Hz, J_2 = 6.4 Hz), 3.29 (6H, s), 4.33 (1H, t, J_1 = 2.9 Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 2.4, 2.8, 17.9, 22.0, 24.5, 25.7, 29.1, 29.37, 29.39, 29.5, 29.7, 32.4, 37.2, 52.5 (2C), 76.9, 104.4. GC/MS: 286 ($M^{+\bullet}$, 0.2), 223 (0.5), 152 (2.3), 109 (2.7), 75 (100). Anal. Calcd. for $C_{17}H_{34}O_3$: C, 71.28; H, 11.96. Found: C, 71.13; H, 11.94.

12-Cyclopropyl-12-hydroxydodecanal (33). Procedure B, 97% yield from **32**. ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.20 (2H, d of d of d of d, J_1 = 22.0 Hz, J_2 = 13.1 Hz, J_3 = 4.9 Hz, J_4 = 4.1 Hz), 0.47 (2H, d of d of d of d, J_1 = 17.2 Hz, J_2 = 12.0 Hz, J_3 = 7.7 Hz, J_4 = 3.9 Hz), 0.85 (1H, d of d of d of d, J_1 = 16.3 Hz, J_2 = 8.4 Hz, J_3 = 5.1 Hz, J_4 = 5.1 Hz), 1.20−1.60 (18H, m), 1.86 (1H, br s), 2.39 (2H, t of d, J_1 = 3.7 Hz, J_2 = 0.9 Hz), 2.81 (1H, d of t, J_1 = 8.4 Hz, J_2 = 6.4 Hz), 9.73 (1H, s). ^{13}C NMR (50 MHz, $CDCl_3$) δ 2.4, 2.8, 18.0, 22.0, 25.7, 29.1, 29.31, 29.37, 29.49, 29.54, 29.7, 37.2, 43.9, 77.2, 203.0. GC/MS: 286 ($M^{+\bullet}$, 0.2), 223 (0.5), 152 (2.3), 109 (2.7), 75 (100). Anal. Calcd. for $C_{15}H_{28}O_2$: C, 74.95; H, 11.74. Found: C, 74.97; H, 12.12.

12-Cyclopropyl-12-hydroxydodecanoic Acid (6a). Procedures E and I, 62% yield from **33**, mp 62.0−63.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.20 (2H, d of d of d of d, J_1 = 22.0 Hz, J_2 = 13.1 Hz, J_3 = 4.9 Hz, J_4 = 4.1 Hz), 0.48 (2H, d of d of d of d, J_1 = 17.2 Hz, J_2 = 12.0 Hz, J_3 = 7.7 Hz, J_4 = 3.9 Hz), 0.87 (1H, d of d of d of d, J_1 = 16.3 Hz, J_2 = 8.4 Hz, J_3 = 5.1 Hz, J_4 = 5.1 Hz), 1.20−1.55 (20H, m), 2.32 (2H, t, J_1 = 7.5 Hz), 2.83 (1H, d of t, J_1 = 8.4 Hz, J_2 = 6.4 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 2.4, 2.8, 17.9, 24.6, 25.6, 28.9, 29.1, 29.3, 29.4, 29.5, 29.6, 34.0, 37.1, 77.1, 179.4. Anal. Calcd. for $C_{15}H_{28}O_3$: C, 70.27; H, 11.01. Found: C, 70.23; H, 11.31.

Methyl 12-Cyclopropyl-12-hydroxydodecanoate (6). Prepared by derivatization of **6a** with an ethereal solution of diazomethane. GC/MS: 237 (0.2), 200 (15.0), 157 (12.9), 143 (17.7), 43 (100).

Methyl 14-Cyclopropyl-14-hydroxytetradec-2-enoate (34). Procedure F, 79% yield from **33**. ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.18 (2H, d of d of d of d, J_1 = 22.0 Hz, J_2 = 13.1 Hz, J_3 = 4.9 Hz, J_4 = 4.1 Hz), 0.46 (2H, d of d of d of d, J_1 = 17.2 Hz, J_2 = 12.0 Hz, J_3 = 7.7 Hz, J_4 = 3.9 Hz), 0.86 (1H, d of d of d of d, J_1 = 16.3 Hz, J_2 = 8.4 Hz, J_3 = 5.1 Hz, J_4 = 5.1 Hz), 1.20−1.45 (16H, m), 1.54 (2H, t, J_1 = 6.7 Hz), 1.66 (1H, br s), 2.15 (2H, d of d of d, J_1 = 7.0 Hz, J_2 = 6.4 Hz, J_3 = 1.5 Hz), 2.80 (1H, d of t, J_1 = 8.5 Hz, J_2 = 6.1 Hz), 3.69 (3H, s), 5.77 (1H, d of t, J_1 = 15.7 Hz, J_2 = 1.5 Hz), 6.94 (1H, d of t, J_1 = 15.7 Hz, J_2 = 6.9 Hz). ^{13}C NMR (50 MHz, $CDCl_3$) δ 2.4, 2.7, 18.0, 25.7, 27.9, 29.0, 29.3, 29.40, 29.43, 29.5, 29.7, 32.2, 37.2, 51.4, 76.9, 120.7, 149.9, 167.2. GC/MS: 247 (0.3), 226 (1.8), 152 (5.4), 129 (14.4), 43 (100). Anal. Calcd. for $C_{18}H_{32}O_3$: C, 71.60; H, 10.52. Found: C, 71.22; H, 10.90.

14-Cyclopropyl-14-hydroxytetradecanoic Acid (8a). Procedures C and G, 55% yield from **34**, mp 78.0−79.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.20 (2H, d of d of d of d, J_1 = 22.0 Hz, J_2 = 13.1 Hz, J_3 = 4.9 Hz, J_4 = 4.1 Hz), 0.48 (2H, d of d of d of d, J_1 = 17.2 Hz, J_2 = 12.0 Hz, J_3 = 7.7 Hz, J_4 = 3.9 Hz), 0.87 (1H, d of d of d of d, J_1 = 16.3 Hz, J_2 = 8.4 Hz, J_3 = 5.1 Hz, J_4 = 5.1 Hz), 1.20−1.45 (20H, m), 1.58 (2H, t of d, J_1 = 6.3 Hz, J_2 = 1.5 Hz), 1.61 (2H, quint, J_1 = 7.3 Hz), 2.32 (2H, t, J_1 = 7.5 Hz), 2.83 (1H, d of t, J_1 = 8.5 Hz, J_2 = 6.4 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 2.4, 2.8, 17.9, 24.6, 25.6, 28.9, 29.1, 29.25, 29.33, 29.40, 29.43, 29.47, 29.6, 34.0, 37.1, 77.1, 179.4. Anal. Calcd. for $C_{17}H_{32}O_3$: C, 71.79; H, 11.34. Found: C, 71.73; H, 11.70.

Methyl 14-Cyclopropyl-14-hydroxytetradecanoate (8). Prepared by derivatization of **8a** with an ethereal solution of diazomethane. GC/MS: 280 (0.2), 228 (16.1), 185 (10.7), 143 (17.4), 43 (100).

1-(Tetrahydropyran-2′-yloxy)-10-(hydroxymethyl)-tetradec-11-ene (35). To a solution of diisopropylamine (355 μL, 2.58 mmol) in THF (10 mL) being stirred at −5 °C under a nitrogen atmosphere was added *n*-butyllithium (1.54 mL, 2.37 mmol, 1.5M in hexanes). After 30 min, *Z*-hex-2-enal-*N*,*N*′-dimethyl hydrazone[30] (0.300 g, 2.15 mmol) was added, and 1 h later, 9-bromo-1-(tetrahydropyran-2′-yloxy)-nonane[49] (0.650 g, 2.37 mmol) was added. The reaction was left to stir for 12 h before being quenched by water (50 mL). $CuCl_2$ (0.310 g, 2.37 mmol) was added, and the reaction was stirred for 1 h. The reaction mixture was then quenched by addition of aqueous, 3 N NH_4OH (30 mL) and extracted with ethyl acetate (3 × 20 mL), and the combined organic layers were washed with brine (30 mL), dried over Na_2SO_4, and concentrated in vacuo. The residue was dissolved in methanol (20 mL) and sodium borohydride (0.050 g, 6.00 mmol) added with stirring under a nitrogen atmosphere. After 2 h, the reaction mixture was quenched by addition of aqueous, 5% oxalic acid (50 mL) and extracted with CH_2Cl_2 (3 × 30 mL). The combined organic layers were washed with brine (30 mL), dried over Na_2SO_4, and concentrated in vacuo. The residue was purified using flash chromatography (silica gel, 10% ethyl acetate in hexanes) to afford **35** (0.135 g, 0.43 mmol, 20% yield over three steps, 75% *E* isomer) as a clear, colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$, *E* and *Z* isomers) δ 0.95 (3H, t, J_1 = 7.5 Hz), 1.20−1.80 (23H, m), 2.06 (2H, m), 2.34 (1H, m), 3.25−3.90 (6H, 4m) 4.54 (1H, t, J_1 = 3.5 Hz), 5.03 (0.75H, d of d of t, J_1 = 21 Hz, J_2 = 8.6 Hz, J_3 = 1.5 Hz), 5.11 (0.25H, d of d of t, J_1 = 8.8 Hz, J_2 = 7.7 Hz, J_3 = 1.5 Hz), 5.56 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* isomer) δ 14.5, 19.7, 21.1, 25.5, 26.2, 27.1, 29.43, 29.47, 29.52, 29.68, 29.72, 30.8, 31.4, 40.4, 62.3, 66.5, 67.7, 98.8, 130.4, 135.2. GC/MS (hydrazone): 366 ($M^{+\bullet}$, 4.1), 337 (2.3), 281 (8.4), 238 (9.8), 139 (64.1), 101 (2.3), 85 (93.0), 41 (100). GC/MS (aldehyde): 324 ($M^{+\bullet}$, 0.8), 294 (0.6), 272 (0.6), 240 (1.3), 123 (1.1), 101 (11.9), 85 (100), 41 (61.8). GC/MS (alcohol): 297 (0.6), 224 (1.1), 149 (1.0), 123 (2.0), 109 (3.6), 101 (8.0), 85 (100), 41 (58.3). Anal. Calcd. for $C_{20}H_{38}O_3$: C, 73.57; H, 11.73. Found: C, 73.37; H, 11.82.

1-(Tetrahydropyran-2′-yloxy)-10-(*t*-butyldiphenylsiloxymethyl)-tetradec-11-ene (36). To a solution of **35** (0.040 g, 0.127 mmol) in DMF (2 mL) being stirred at room temper-

(49) Sharma, A.; Chattopadhyay, S. *J. Org. Chem.* **1998**, *63*, 6128−6131.

ature was added TBDPSCl (43 μL, 0.153 mmol) and imidazole (0.020 g, 0.254 mmol). After 10 min, the reaction was quenched by the addition of water (10 mL)/diethyl ether (10 mL), and the aqueous phase was extracted with diethyl ether (10 mL). The combined organic layers were washed with brine (20 mL), dried over Na_2SO_4, and concentrated in vacuo. The residue was purified using flash chromatography (silica gel, 5% ethyl acetate in hexanes) to afford **36** (0.0453 g, 0.0828 mmol, 65% yield, 75% *E* isomer) as a clear, colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$, *E* and *Z* isomers) δ 0.90 (3H, t, J_1 = 7.5 Hz), 1.04 (9H, s), 1.20−1.60 (20H, m), 1.70−1.85 (2H, 2m), 1.95 (2H, m), 2.52 (1H, m), 3.35−3.90 (6H, 4m) 4.57 (1H, t, J_1 = 2.7 Hz), 5.06 (0.75H, d of d of t, J_1 = 21.0 Hz, J_2 = 8.4 Hz, J_3 = 1.5 Hz), 5.11 (0.25H, d of d of t, J_1 = 8.8 Hz, J_2 = 8.3 Hz, J_3 = 1.5 Hz), 5.56 (1H, m), 7.38 (6H, m), 7.66 (4H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* isomer) δ 14.5, 19.7 (3C), 21.0, 25.5, 26.2, 26.9, 27.1, 29.49, 29.53, 29.6, 29.68, 29.72, 29.8, 30.8, 31.6, 40.2, 62.3, 66.6, 67.7, 98.8, 127.5 (4C), 129.4 (2C), 131.0, 132.8, 134.0 (2C), 135.6 (4C). Anal. calcd. for $C_{36}H_{56}O_3$: C, 76.55; H, 9.99. Found: C, 76.33; H, 10.21.

Methyl 10-(*t*-Butyldiphenylsiloxymethyl)-tetradec-11-enoate (37). Procedures B and E, 80% yield from **36**, 75% *E* isomer. ^{1}H NMR (400 MHz, $CDCl_3$, *E* and *Z* isomers) δ 0.89 (3H, t, J_1 = 7.5 Hz), 1.03 (9H, s), 1.10−1.70 (14H, m), 1.97 (2H, m), 2.28 (2H, t, J_1 = 7.6 Hz), 2.50 (1H, m), 3.48 (2H, d, J_1 = 6.4 Hz), 3.65 (3H, s), 5.05 (0.75H, m), 5.19 (0.25H, m), 5.42 (1H, m), 7.38 (6H, m), 7.66 (4H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* isomer) δ 14.5, 19.3 (3C), 21.0, 25.0, 26.9, 27.0, 29.1, 29.4, 29.7, 30.3, 31.6, 34.1, 40.0, 51.4, 67.6, 127.5 (4C), 129.5 (2C), 130.9, 132.9, 134.0 (2C), 135.6 (4), 174.3. Anal. Calcd. for $C_{32}H_{48}O_3Si$: C, 75.54; H, 9.51. Found: C, 75.40; H, 9.58. HRMS: calcd for $C_{32}H_{48}O_3SiNa$, 531.3271; found, 531.3273.

Methyl 10-Hydroxymethyltetradec-11-enoate (11). To a solution of **37** (0.015 g, 0.0308 mmol) in THF (20 mL) being stirred at 0 °C was added TBAF (23 μL, 1.0 M in THF, 0.0370 mmol) and acetic acid (1 μL, 0.0308 mmol), and the reaction was stirred at room temperature for 30 h before being quenched by addition of brine (20 mL). Following separation of the layer, the aqueous phase was extracted with diethyl ether (3 × 10 mL), the combined organic layers were dried over Na_2SO_4, and the solvent was removed in vacuo. The residue was purified using flash chromatography (silica gel, 10% ethyl acetate in hexanes) to afford **11** (0.008 g, 0.0308 mmol, 100% yield, 75% *E* isomer) as a clear, colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$, *E* and *Z* isomers) δ 0.95 (3H, t, J_1 = 7.5 Hz), 1.20−1.60 (15H, m), 2.06 (2H, m), 2.28 (2H, t, J_1 = 7.5 Hz), 2.57 (1H, m), 3.30, (1H, t, J_1 = 9.3 Hz), 3.52 (1H, m), 3.64 (3H, s), 5.01 (0.75H, d of d of t, J_1 = 21.0 Hz, J_2 = 8.4 Hz, J_3 = 1.5 Hz), 5.11 (0.25H, d of d of t, J_1 = 8.8 Hz, J_2 = 8.3 Hz, J_3 = 1.5 Hz), 5.59 (1H, d of t, J_1 = 11.7 Hz, J_2 = 7.4 Hz). ^{13}C NMR (100 MHz, $CDCl_3$, *E* isomer) δ 14.5, 21.1, 24.9, 27.1, 29.1, 29.2, 29.3, 29.6, 31.4, 34.1, 40.4, 51.4, 66.5, 134.0, 135.6, 174.3. GC/MS: 252 ($M^{+\bullet}$ − H_2O, 2.0), 240 (8.5), 208 (23.0), 166 (16.4), 123 (16.3), 109 (22.9), 69 (70.7), 55 (100). Anal. Calcd. for $C_{16}H_{30}O_3$: C, 71.07; H, 11.18. Found: C, 71.07; H, 11.18.

10-(Tetrahydropyran-2′-yloxy)-decanal (38). Procedure K, 75% yield from 1-(tetrahydropyran-2′-yloxy)-undec-10-ene.[18] ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.20−1.80 (22H, m), 3.30−3.90 (4H, 4m), 4.53 (1H, t, J_1 = 3.5 Hz), 9.72 (1H, t, J_1 = 1.8 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 19.6, 22.0, 25.4, 26.1, 29.2, 29.3, 29.4, 29.5, 29.6, 30.7, 43.8, 62.3, 67.6, 98.8, 202.9. GC/MS: 256 ($M^{+\bullet}$, 0.1), 255 (0.7), 183 (0.5), 155 (1.1), 137 (1.8), 101 (35.2), 85 (100), 41 (71.9). Anal. Calcd. for $C_{15}H_{28}O_3$: C, 70.27; H, 11.01. Found: C, 70.10; H, 11.25.

1-(Tetrahydropyran-2′-yloxy)-tridec-12-en-10-ol (39). Procedure J, 87% yield from **38**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.20−1.80 (23H, m), 2.02−2.28 (2H, 2m), 3.30−3.50 (2H, 2m), 3.57 (1H, t of t, J_1 = 6.7 Hz, J_2 = 5.4 Hz), 3.65−3.85 (2H, 2m), 4.53 (1H, t, J_1 = 3.5 Hz), 5.06 (1H, m), 5.09 (1H, m), 5.79 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$) δ 19.6, 25.4, 25.6, 26.1, 29.36, 29.43, 29.45, 29.54, 29.7, 30.7, 36.7, 41.9, 62.2, 67.6, 70.6, 98.7, 117.8, 134.9. GC/MS: 297 (0.2), 281 (0.5), 173 (1.9), 137 (2.6), 109 (2.1), 101 (12.9), 85 (98.9), 41 (100). Anal. Calcd. for $C_{18}H_{34}O_3$: C, 72.44; H, 11.48. Found: C, 72.59; H, 11.59.

1-(Tetrahydropyran-2′-yloxy)-10-hydroxypentadec-12-ene (40). Procedures K and L, 72% yield from **39**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.95 (3H, t, J_1 = 7.5 Hz), 1.20−1.85 (23H, m), 2.05 (2H, m), 2.19 (2H, m), 3.30−3.50 (2H, 2m), 3.55 (1H, m), 3.65−3.85 (2H, 2m), 4.55 (1H, t, J_1 = 3.5 Hz), 5.36 (1H, m), 5.56 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$) δ 14.2, 19.7, 20.7, 25.5, 25.7, 26.2, 29.4, 29.5, 29.6, 29.7, 30.8, 35.2, 36.7, 40.6, 62.3, 67.7, 71.5, 98.8, 124.5, 135.1. GC/MS: 283 (0.2), 224 (0.7), 173 (4.0), 109 (1.9), 101 (10.7), 85 (100), 43 (23.7), 41 (38.7). Anal. Calcd. for $C_{20}H_{38}O_3$: C, 73.57; H, 11.73. Found: C, 73.77; H, 11.67.

Pentadec-12-en-1,10-diol (41). Procedure B, 90% yield from **40**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.95 (3H, t, J_1 = 7.5 Hz), 1.20−1.60 (18H, m), 2.05 (2H, m), 2.20 (2H, m), (3.68, 1H, quint, J_1 = 5.8 Hz), 3.62 (2H, t, J_1 = 6.6 Hz), 5.36 (1H, m), 5.55 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$) δ 14.2, 20.7, 25.7, 29.4, 29.5, 29.6, 29.7, 32.8, 35.2, 36.8, 40.6, 63.1, 71.5, 124.5, 135.1. Anal. Calcd. for $C_{15}H_{30}O_2$: C, 74.33; H, 12.41. Found: C, 74.27; H, 12.10.

Methyl 10-Oxopentadec-12-enoate (42). Procedure E, esterification with ethereal diazomethane, 88% yield from **41**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.95 (3H, t, J_1 = 7.5 Hz), 1.20−1.60 (12H, m), 2.03 (2H, quint, J_1 = 7.4 Hz), 2.27 (2H, t, J_1 = 7.9 Hz), 2.39 (2H, t, J_1 = 7.4 Hz), 3.12 (2H, d, J_1 = 6.9 Hz), 3.64 (3H, s), 5.51 (2H, m). ^{13}C NMR (100 MHz, $CDCl_3$) δ 13.9, 20.8, 23.7, 24.9, 29.05, 29.11, 29.2, 29.7, 34.1, 41.6, 42.3, 51.4, 121.0, 135.2, 174.3, 209.3. GC/MS: 268 ($M^{+\bullet}$, 1.0), 237 (3.5), 200 (6.0), 199 (49.9), 139 (29.8), 121 (21.8), 41 (100). Anal. Calcd. for $C_{16}H_{28}O_3$: C, 71.60; H, 10.51. Found: C, 71.44; H, 10.74.

Methyl 10-Hydroxypentadec-12-enoate (13). Procedure I, 75% yield from **42**, mp 34.0−35.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.95 (3H, t, J_1 = 7.5 Hz), 1.20−1.60 (15H, m), 2.03 (2H, d of t, J_1 = 7.4 Hz, J_2 = 6.0 Hz), 2.18 (2H, d of t, J_1 = 7.3 Hz, J_2 = 6.3 Hz), 2.27 (2H, t, J_1 = 7.5 Hz), 3.58 (1H, m), 3.64 (3H, s), 5.36 (1H, m), 5.55 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$) δ 14.2, 20.7, 24.9, 25.7, 29.1, 29.2, 29.6, 34.1, 35.2, 36.7, 40.7, 51.4, 71.4, 124.5, 135.1, 174.3. GC/MS: 252 (0.3), 201 (15.4), 170 (8.5), 169 (78.8), 151 (4.7), 133 (8.5), 55 (98.7), 41 (100). Anal. Calcd. for $C_{16}H_{30}O_3$: C, 71.07; H, 11.18. Found: C, 70.93; H, 11.44.

1-(Tetrahydropyran-2′-yloxy)-tridec-10-en-13-ol (43). Procedure L, 86% yield from **38**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.20−1.80 (21H, m), 1.98 (1H, q, J_1 = 6.9 Hz), 2.03 (1H, q, J_1 = 6.8 Hz), 2.23 (1H, q of q, J_1 = 7.0 Hz, J_2 = 1.0 Hz), 2.30 (1H, q of q, J_1 = 7.2 Hz, J_2 = 0.6 Hz), 3.30−3.50 (2H, 2m), 3.60 (2H, br d, J_1 = 4.4 Hz), 3.65−3.90 (2H, 2m), 4.55 (1H, t, J_1 = 3.5 Hz), 5.35 (1H, m), 5.52 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* & *Z* isomers) δ 19.7, 25.5, 26.2, 27.3, 29.1, 29.38, 29.40, 29.42, 29.5, 29.6, 29.7, 30.76, 30.78, 36.0, 62.0, 62.3, 67.7, 98.8, 124.9, 125.7, 133.5, 134.3. GC/MS: 297 (0.2), 268 (0.5), 196 (1.3), 137 (1.0), 101 (15.0), 85 (100), 55 (34.2), 41 (48.8). Anal. Calcd. for $C_{18}H_{34}O_3$: C, 72.44; H, 11.48. Found: C, 72.31; H, 11.59.

13-(Tetrahydropyran-2′-yloxy)-tridec-3-enal (44). Procedure H, 94% yield from **43**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.20−1.80 (20H, m), 2.02 (2H, 2t, J_1 = 7.0 Hz), 3.08 (1H, d of m, J_1 = 7.2 Hz), 3.16 (1H, d of m, J_1 = 7.2 Hz), 3.30−3.90 (4H, 4m), 4.55 (1H, t, J_1 = 3.5 Hz), 5.42−5.72 (2H, m), 9.63 (1H, d of t, J_1 = 2.5 Hz, J_2 = 2.0 Hz). ^{13}C NMR (100 MHz, $CDCl_3$, *E* and *Z* isomers) δ 19.7, 25.5, 26.2, 27.6, 29.1, 29.39, 29.41, 29.44, 29.5, 29.7, 30.8, 32.7, 42.6, 47.3, 62.4, 67.7, 98.9, 117.9, 119.0, 135.5, 137.0, 199.8, 200.4. GC/MS: 296 ($M^{+\bullet}$, 0.1), 177 (0.4), 135 (1.7), 121 (4.0), 101 (53.5), 85 (100), 55 (25.5), 41 (67.2). Anal. Calcd. for $C_{18}H_{32}O_3$: C, 72.93; H, 10.88. Found: C, 72.55; H, 10.80.

1-(Tetrahydropyran-2′-yloxy)-tetradec-10-en-13-ol (45). Procedure J, 87% yield from **44**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.15 (1.5H, d, J_1 = 6.2 Hz), 1.18 (1.5H, d, J_1 = 6.2 Hz), 1.20−1.80 (21H, m), 1.95−2.25 (4H, 2m), 3.30−3.50 (2H, 2m), 3.65−

3.85 (3H, 3m), 4.55 (1H, t, J_1 = 3.5 Hz), 5.38 (1H, m), 5.55 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* and *Z* isomers) δ 19.7, 22.6, 22.7, 25.5, 26.2, 27.4, 29.1, 29.3, 29.39, 29.41, 29.43, 29.5, 29.7, 30.8, 32.6, 37.1, 42.5, 62.3, 67.2, 67.7 (2C), 98.8, 125.0, 125.7, 133.5, 134.8. GC/MS: 281 (0.5), 210 (1.1), 195 (1.0), 123 (1.7), 101 (13.3), 85 (100), 45 (35.9), 41 (36.3). Anal. Calcd. for $C_{19}H_{36}O_3$: C, 73.03; H, 11.61. Found: C, 73.06; H, 11.30.

Tetradec-10-en-1,13-diol (46). Procedure B, 91% yield from **45**. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.16 (1.5H, d, J_1 = 6.2 Hz), 1.19 (1.5H, d, J_1 = 6.2 Hz), 1.20−1.60 (16H, m), 1.98−2.10 (3H, m), 2.30 (1H, m), 3.62 (2H, t, J_1 = 6.6 Hz), 3.78 (1H, m), 5.38 (1H, m), 5.55 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* & *Z* isomers) δ 22.6, 22.8, 25.7, 27.4, 29.1, 29.2, 29.36, 29.41, 29.48, 29.50, 29.6, 29.7, 32.6, 32.8, 37.1, 42.5, 63.1, 67.2, 67.7, 125.0, 125.7, 133.5, 134.8. Anal. Calcd. For $C_{14}H_{28}O_2$: C, 73.63; H, 12.36. Found C, 73.72; H, 12.23.

(2-Hydroxypropanol)-9-cyclopropylnonan-1-ol (47). Procedure D, 89% yield from **46**. ^{1}H NMR (400 MHz, $CDCl_3$) δ −0.25 (0.5H, m), 0.22 (1H, m), 0.43 (1H, m), 0.68 (1.5H, m), 1.18 (1.5H, d, J_1 = 6.4 Hz), 1.22 (1.5H, d, J_1 = 6.4 Hz), 1.25−1.70 (20H, m), 3.61 (2H, t, J_1 = 6.6 Hz), 3.86 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* and *Z* isomers) δ 10.9, 11.7, 12.3, 14.7, 15.1, 15.2, 18.1, 23.1, 25.7, 28.9, 29.37, 29.39, 29.44, 29.5, 29.55, 29.56, 29.7, 30.0, 32.8, 38.0, 38.1, 43.7, 63.1, 68.8, 69.1. HRMS: calcd for $C_{15}H_{30}O_2$, 242.2246; found, 242.2248. Anal. Calcd. for $C_{15}H_{28}O_2$: C, 74.32; H, 12.47. Found: C, 74.67; H, 12.10.

Methyl (2-Hydroxypropanol)-9-cyclopropylnonanoate (9). Procedure E, esterification with ethereal diazomethane, procedure I, 90% yield from **47**, mp 39.0−40.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$, *E* and *Z* isomers) δ −0.25 (0.5H, m), 0.22 (1H, m), 0.43 (1H, m), 0.68 (1.5H, m), 1.18 (1.5H, d, J_1 = 6.4 Hz), 1.22 (1.5H, d, J_1 = 6.4 Hz), 1.25−1.80 (17H, m), 2.28 (2H, t, J_1 = 7.5 Hz), 3.64 (3H, s), 3.87 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E* and *Z* plus syn/anti isomers) δ 10.8, 10.9, 11.2, 11.7, 12.2, 12.3, 14.7, 15.1, 15.2, 15.4, 18.1, 18.7, 23.1, 23.3, 24.9, 28.9, 29.0, 29.1, 29.2, 29.4, 29.5, 29.6, 29.7, 30.0, 34.10, 34.11, 34.2, 38.0, 38.1, 43.70, 43.72, 51.4, 68.6, 68.75, 68.81, 69.0, 174.4. GC/MS: 270 ($M^{+\bullet}$, 0.1), 252 (0.4), 220 (1.0), 194 (2.3), 178 (1.7), 152 (3.5), 55 (67.3), 45 (100). Anal. Calcd. for $C_{16}H_{30}O_3$: C, 71.07; H, 11.18. Found: C, 70.89; H, 11.21.

Methyl 15-Hydroxypentadec-12-ene (15). Procedure L, 60% yield from methyl 12-oxododecanoate,[32] 75% *E* isomer, mp 44.0−45.0 °C. ^{1}H NMR (400 MHz, $CDCl_3$, *E* isomer) δ 1.20−1.35 (14H, m), 1.57 (2H, quint, J_1 = 7.5 Hz), 1.68 (1H, br s), 1.95 (2H, d of t, J_1 = 6.9 Hz, J_2 = 6.7 Hz), 2.22 (2H, d of d of t, J_1 = 6.0 Hz, J_2 = 0.90 Hz, J_3 = 7.4 Hz), 2.25 (2H, t, J_1 = 7.5 Hz), 3.57 (2H, t, J_1 = 6.4 Hz), 3.61 (3H, s), 5.33 (1H, m), 5.49 (1H, m). ^{13}C NMR (100 MHz, $CDCl_3$, *E*-isomer) δ 24.9, 29.07, 29.16, 29.19, 29.33, 29.37, 29.44, 29.6, 30.7, 34.0, 35.9, 51.4, 62.0, 125.7, 134.2, 174.3. GC/MS: 252 (1.6), 240 (2.7), 208 (9.6), 166 (6.8), 124 (8.5), 110 (11.2), 55 (94.0), 41 (100). Anal. Calcd. for $C_{16}H_{30}O_3$: C, 71.07; H, 11.18. Found: C, 70.73; H, 11.54.

Acknowledgment. M.J.C. is grateful to the University of Queensland Graduate School for the Graduate School Travel Award and the University of Queensland Department of Chemistry for the Francine Kroesen Scholarship.

Supporting Information Available: General methods and instrumentation; experimental procedures for **1a**, **2a**, ***E*-3a**, **4a**−**8a**, **1**−**9**, **13**, **15**, **19**, ***Z*-20**, **21**, **23**−**27**, ***Z*-28**, **29**, **31**−**34**, and **37**−**47**; GC conditions for turnover analysis; retention times of synthesized standards; and NMR determination of rearrangement product from ***E*-1a**. This information is available free of charge via the Internet at http://pubs.acs.org.

JO047985D