Journal of Medicinal Chemistry

ARTICLE

pubs.acs.org/jmc

A Prodrug Approach for Improving Antituberculosis Activity of Potent *Mycobacterium tuberculosis* Type II Dehydroquinase Inhibitors[†,‡]

Lorena Tizón,[§] José M. Otero,[∥] Verónica F. V. Prazeres,[§] Antonio L. Llamas-Saiz,[⊥] Gavin C. Fox,[∥] Mark J. van Raaij,[#] Heather Lamb,[▽] Alastair R. Hawkins,[▽] José A. Ainsa,[○] Luis Castedo,[◆] and Concepción González-Bello*[,§]

[§]Centro Singular de Investigación en Química Biológica y Materiales Moleculares, Universidad de Santiago de Compostela, calle Jenaro de la Fuente s/n, 15782 Santiago de Compostela, Spain

[∥]Laboratoire des Proteines Membranaires, Institut de Biologie Structurale J. P. Ebel, 38027 Grenoble, France

[⊥]Unidad de Rayos X, RIAIDT, Edificio CACTUS, Universidad de Santiago de Compostela, 15782 Santiago de Compostela, Spain

[#]Departamento de Estructura de Macromoléculas, Centro Nacional de Biotecnología (CSIC), Campus Cantoblanco, 28049 Madrid, Spain

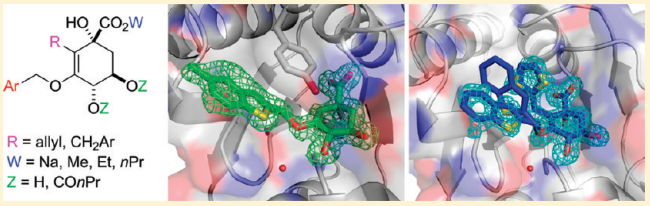
[▽]Institute of Cell and Molecular Biosciences, Medical School, University of Newcastle upon Tyne, Newcastle upon Tyne NE2 4HH, U.K.

[○]Grupo de Genética de Micobacterias, Departamento de Microbiología, Medicina Preventiva y Salud Pública, Facultad de Medicina, 50009 Zaragoza, Spain, and CIBER Enfermedades Respiratorias, Spain

[◆]Departamento de Química Orgánica, Facultad de Química, Universidad de Santiago de Compostela, Avenida de las Ciencias s/n, 15782 Santiago de Compostela, Spain

Supporting Information

ABSTRACT: The synthesis of high-affinity reversible competitive inhibitors of *Mycobacterium tuberculosis* type II dehydroquinase, an essential enzyme in *Mycobacterium tuberculosis* bacteria, is reported. The inhibitors reported here are mimics of the enol intermediate and the effect of substitution on C2 was studied. The crystal structures of *Mycobacterium tuberculosis* type II dehydroquinase in complex with three of the reported inhibitors are also described. The results show that an aromatic substituent on C2 prevents the closure of the active site by impeding the hydrogen-bonding interaction of Arg108 with the essential Tyr24 of the flexible loop, the residue that initiates catalysis. Chemical modifications of the reported acids were also carried out to improve internalization into *Mycobacterium tuberculosis* through an ester prodrug approach. Propyl esters proved to be the most efficient in achieving optimal in vitro activities.



INTRODUCTION

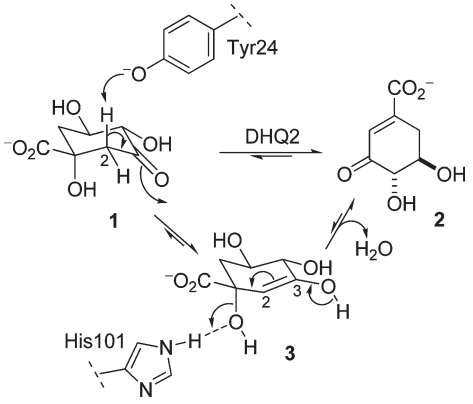
Tuberculosis (TB), which is caused by *Mycobacterium tuberculosis*, remains one of the deadliest infectious diseases for humans, and it has been identified by the World Health Organization (WHO) as one of the three priority diseases for drug research and development.[1] Thanks to global control efforts over the past decade, the estimated worldwide incidence rate fell to 137 cases per 100000 people in 2009, after peaking in 2004 at 142 cases per 100000.[1] However, the rate is still falling too slowly.

The concomitant emergence of HIV and the surge of multidrug-resistant isolates of *M. tuberculosis* (MDR-TB) have reaffirmed tuberculosis as a primary public health threat.[2] In 2010, the largest WHO MDR-TB survey reported the highest-ever rates of MDR-TB, with peaks of up to 28% of new TB cases in some regions of the former Soviet Union. The continued evolution and aggravation of the drug resistance problem has led to extensively drug-resistant TB (XDR-TB), which is resistant to first-line and second-line TB antibiotics.[3,4] MDR and XDR forms of TB are extraordinarily difficult to treat, particularly in patients with a compromised immune system.[5]

TB therapy requires long periods of treatment (around 6−24 months) with multiple drugs. This extended treatment leads to poor compliance, a factor that promotes resistance development. The various antibiotics that constitute the first- and second-line drugs for TB therapy target only a small number of essential functions of the bacterium. It is therefore necessary discover further pathways that are required for bacterial growth and to develop compounds that target these pathways.[6−8] This approach should provide novel targets for the rational design of more effective treatments for tuberculosis, which should also reduce the duration of therapy necessary to eradicate *M. tuberculosis* from the patient.

Received: May 13, 2011
Published: July 22, 2011

Scheme 1. Proposed E1CB Mechanism for the Enzymatic Conversion of 3-Dehydroquinic Acid (1) to 3-Dehydroshikimic Acid (2) Catalyzed by DHQ2[a]



[a] The reaction proceeds via the enol intermediate **3**. Relevant residues are indicated.

The shikimate pathway is an attractive target for antibiotic development because it is present in bacteria, fungi, plants, and in apicomplexan parasites such as *Plasmodium falciparum* (malaria),[9] *Toxoplasma gondii*, and *Cryptosporidium parvum*, but it is absent in mammals. This route involves seven enzymes that catalyze the sequential conversion of erythrose-4-phosphate and phosphoenol pyruvate to chorismic acid, an essential precursor in the synthesis of aromatic amino acids and other metabolites, including folate cofactors, ubiquinone, and vitamins E and K.[10] The absence of the shikimate pathway in mammals, combined with its essential nature in *M. tuberculosis*,[11] makes it an attractive target for the development of new anti-TB agents.

Dehydroquinase (3-dehydroquinate dehydratase, DHQ, EC 4.2.1.10) is the third enzyme of the shikimate pathway, and it catalyzes the reversible dehydration of 3-dehydroquinic acid (**1**) to form 3-dehydroshikimic acid (**2**) (Scheme 1). This reaction is part of two metabolic pathways: the biosynthetic shikimate pathway and the catabolic quinate pathway. There are two distinct types of enzymes, known as type I (DHQ1) and type II (DHQ2), which have different biochemical and biophysical properties and do not show any sequence similarity. DHQ1, which is found in plants, fungi, and many bacterial species (for example *Salmonella typhi* and *Escherichia coli*), is exclusively biosynthetic, whereas DHQ2 (found in *Streptomyces coelicolor, Mycobacterium tuberculosis, Aspergillius nidulans, Helicobacter pylori*, and *Neurospora crassa*) has both biosynthetic and catabolic roles. DHQ1 and DHQ2 utilize completely different mechanisms to catalyze the same overall reaction.[12] The type I mechanism involves covalent imine intermediates between the enzyme and the substrate and proceeds with *syn* stereochemistry.[13] In contrast, the type II reaction proceeds through an enol intermediate with overall *anti* stereochemistry (Scheme 1).[14] The reaction is initiated by an essential tyrosine of the active site, which removes the *pro*-S hydrogen from C2 of **1**. The final step is the acid-catalyzed elimination of the C-1 hydroxyl group, a reaction mediated by His101 acting as a proton donor. Two residues, Arg19 and Tyr24, have been identified by chemical modification and site-directed mutagenesis studies as being essential for enzyme activity.[15] Both residues reside in a flexible loop that closes over the active site upon substrate binding. The essential arginine of the loop is presumed to orient the tyrosine in an appropriate manner for proton abstraction.[16,17] The proximity of both Arg108 and Arg19 lowers the pK_a of the catalytic tyrosine. It has also been suggested that the essential Arg19 and a conserved water molecule close to the carbonyl group of Asn12 stabilize the enol intermediate.

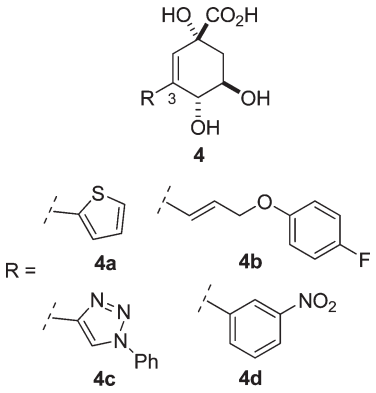


Figure 1. Relevant examples of competitive reversible inhibitors of DHQ2-Mt.

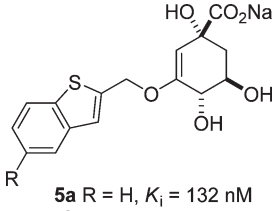
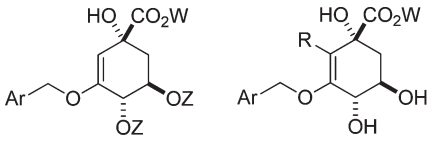


Figure 2. 3-Methoxybenzothiophenyl derivatives that are inhibitors of DHQ2-Hp.

In recent years, we and other groups have designed inhibitors of DHQ2, especially from *M. tuberculosis* (DHQ2-Mt) and *H. pylori* (DHQ2-Hp), two pathogenic bacteria in which the enzyme is essential (the respective genes are *Aro*Q and *Aro*D). Most of the reported inhibitors are mimics of the enol intermediate **3** (compounds **4**, Figure 1).[18−31] Some examples of high-affinity inhibitors of DHQ2-Mt include 2-thienyl compound **4a**[27] (K_i of 250 nM), alkenyl derivative **4b**[26] (K_i of 120 nM), 3-nitrophenyl derivative **4d**[22,31] (K_i of 54 nM), and triazole **4c**[30] (K_i of 39 nM). On the other hand, we recently showed that 3-methoxyaryl derivatives **5a** (K_i of 132 nM) and **5b** (K_i of 130 nM) are potent competitive inhibitors of DHQ2-Hp and the crystal structure of DHQ2-Hp in complex with compound **5b** has been solved at 2.95 Å (Figure 2).[28]

The resolution of this crystal structure has been very important to clarify the role of the aromatic rings on C3 for inhibition. These rings expel the essential arginine side chain from the active site. In fact, this structure, along with others solved later, reveals an important change in the conformation and flexibility of the loop that closes over the substrate binding site.[17,28,29] Molecular dynamics simulation studies suggest that the aromatic ring prevents appropriate orientation of the catalytic tyrosine of the loop for proton abstraction and disrupts its basicity.[17] The synthesis of compounds **5a** and **5b** involved *O*-alkylation, but some *C*-alkylation at C2 was also obtained. It was found that the resulting disubstituted compounds are also potent inhibitors of DHQ2. To investigate in more detail the effect of substitution of

dx.doi.org/10.1021/jm2006063 |*J. Med. Chem.* 2011, 54, 6063–6084



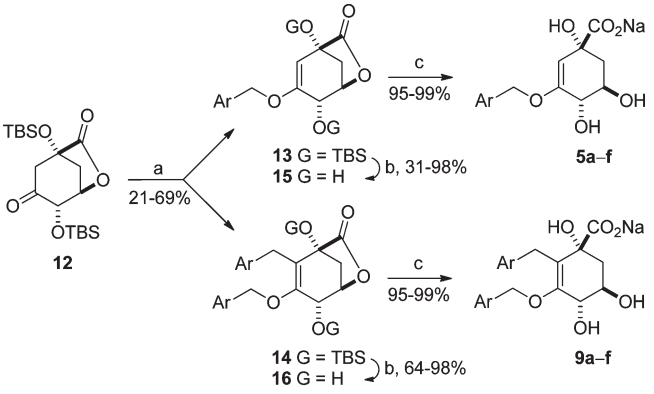
5 W = Na
6 W = Me
7 W = Et
8 W = *n*Pr

9 W = Na
10 W = Me
11 W = Et

Ar = benzo[*b*]thiophen-2-yl,
5-methylbenzo[*b*]thiophen-2-yl,
6-chlorobenzo[*b*]thiophen-2-yl,
benzo[*b*]thiophen-5-yl,
naphth-2-yl,
thien-2-yl,
R = allyl, CH_2Ar
Z = H, CO*n*Pr

Figure 3. Target compounds **5**−**11**.

Scheme 2[a]



a Ar = benzo[*b*]thiophen-2-yl
b Ar = 5-methylbenzo[*b*]thiophen-2-yl
c Ar = 6-chlorobenzo[*b*]thiophen-2-yl
d Ar = benzo[*b*]thiophen-5-yl
e Ar = naphth-2-yl
f Ar = thien-2-yl

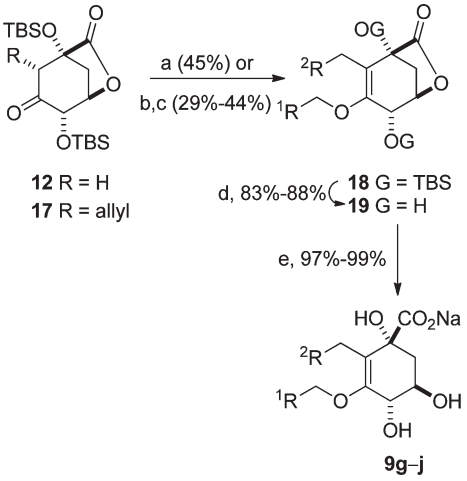
[a] Reagents and conditions: (a) (1) KHMDS, DMF/PhMe, −78 °C, (2) $ArCH_2Br$; (b) TBAF, THF, 0 °C; (c) NaOH, THF, RT.

C2, and therefore the potential for the development of new enzyme inhibitors, we report the synthesis of several 3-substituted and 2,3-disubstituted derivatives, compounds **5** and **9**, respectively (Figure 3). It was found that these compounds are new high affinity inhibitors of DHQ2-Mt. The crystal structure of DHQ2-Mt in complex with compound **5b** and with the doubly substituted enol mimics **9d** and **9h** is described along with the results of inhibition studies on compounds **5** and **9** against DHQ2-Mt and DHQ2-Hp. In addition, chemical modification of the reported acids was carried out with the aim of obtaining improved internalization into *M. tuberculosis*. This resulted in the synthesis of esters **6**−**8** and **10**−**11**. The results of inhibition studies of these comporids against DHQ2-Mt and DHQ2-Hp and the molecular docking studies using GOLD 5.0 are also described.

RESULTS AND DISCUSSION

Synthesis of *O*-Alkylaryl Derivatives 5a−f and Disubstituted Compounds 9a−f. The synthesis of *O*-alkylaryl derivatives **5a**−**f** and dialkyl analogues **9a**−**f** was carried out first. These

Scheme 3[a]



g ^{1}R = benzo[*b*]thiophen-2-yl; ^{2}R = vinyl
h ^{1}R = benzo[*b*]thiophen-2-yl; ^{2}R = thien-2-yl
i ^{1}R = thien-2-yl; ^{2}R = 5-methylbenzo[*b*]thiophen-2-yl
j ^{1}R = thien-2-yl; ^{2}R = benzo[*b*]thiophen-5-yl

[a] Reagents and conditions: (a) (1) KHMDS, DMF/PhMe, −78 °C, (2) $ArCH_2Br$; (b) (1) LiHMDS, THF, RT, (2) $^2RCH_2I$; (c) (1) KHMDS, DMF/PhMe, −78 °C, (2) $^1RCH_2Br$; (d) TBAF, THF, 0 °C; (e) NaOH, THF, RT.

compounds incorporate the same aromatic ring in both positions 2 and 3. The effect of substitution with naphthyl, thienyl, and benzo[*b*]thiophenyl rings was investigated.[23,27,28] In the case of the benzo[*b*]thiophenyl substituent, we further studied the effect of the incorporation of a methyl group and a chloro-substituent. The synthesis of compounds **5a**−**f** and **9a**−**f** was achieved by alkylation of ketone **12**[22] (Scheme 2). Treatment of ketone **12** with KHMDS at −78 °C followed by reaction with the corresponding bromide afforded a chromatographically separable mixture of *O*-alkylated **13a**−**f** and the dialkyl products **14a**−**f**. Finally, deprotection of the TBS groups in lactones **13a**−**f** and **14a**−**f** with TBAF, followed by basic hydrolysis of the corresponding lactones **15a**−**f** and **16a**−**f**, gave the desired enol mimics **5a**−**f** and **9a**−**f**, respectively.

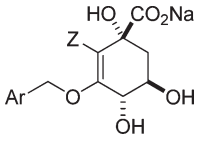
Synthesis of Disubstituted Derivatives 9g−j. To study the relative effect of the type and size of the substituent in position 2, we synthesized compounds **9g**−**j** (Scheme 3). Another reason to synthesize allyl derivative **9g** was to understand the role of an aromatic group on C2.

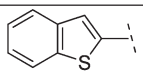
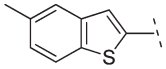
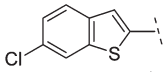
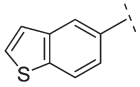
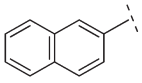
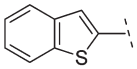
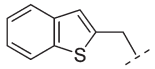
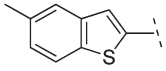
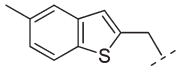
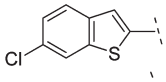
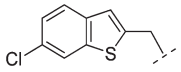
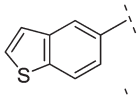
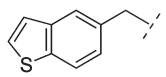
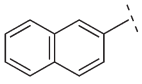
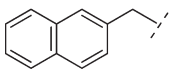
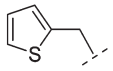
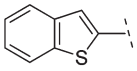
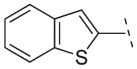
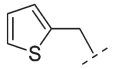
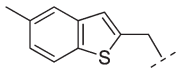
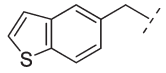
2-Allyl compound **9g** was synthesized in a three-step reaction sequence from our previously reported (2*S*)-2-allyl ketone 17.[32] First, alkylation of ketone **17** by treatment with KHMDS followed by reaction with 2-(bromomethyl)benzo[*b*]thiophene[32] afforded ether **18g** in 45% yield. Finally, compound **18g** was efficiently converted into the 2-allyl derivative **9g** as for compounds **5**.

The synthesis of compounds **9h**−**j** involved the sequential *C*-alkylation and *O*-alkylation of ketone **12** with two different alkylating agents. First, *C*-alkylation of ketone **12** was achieved by treatment of **12** with LiHMDS at room temperature followed by reaction with the appropriate iodide to afford the corresponding 2-alkyl ketones as a mixture of diastereoisomers in C2. Subsequent treatment of the resulting 2-alkyl ketones with KHMDS at −78 °C followed by reaction with the corresponding

dx.doi.org/10.1021/jm2006063 |*J. Med. Chem.* 2011, 54, 6063–6084

Table 1. K_i (nM) of Compounds 5 and 9 against DHQ2-Mt and DHQ2-Hp



Entry	Compound	Ar	Z	DHQ2-Mt[a]	DHQ2-Hp[b]
1	**5a**		H	28 ± 2	132 ± 13[28]
2	**5b**		H	42.5 ± 6	130 ± 13[28]
3	**5c**		H	35 ± 2	205 ± 15
4	**5d**		H	31 ± 3	166 ± 15[28]
5	**5e**		H	35 ± 2	310 ± 46
6	**5f**		H	235 ± 12	920 ± 90[28]
7	**9a**			40 ± 3	97 ± 8
8	**9b**			188 ± 19	50 ± 3
9	**9c**			1100 ± 66	6700 ± 603
10	**9d**			440 ± 40	140 ± 3
11	**9e**			6000 ± 300	260 ± 18
12	**9f**			5100 ± 153	14600 ± 292
13	**9g**		allyl	1240 ± 50	1100 ± 11
14	**9h**			250 ± 12	280 ± 11
15	**9i**			870 ± 17	279 ± 8
16	**9j**			59 ± 5	100 ± 5

[a] 50 mM Tris.HOAc, pH = 7.0, 25 °C. [b] 50 mM Tris.HCl, pH = 7.0, 25 °C.

bromide gave the desired disubstituted compounds **18h**−**j** in low to moderate overall yield. Finally, compounds **18h**−**j** were converted into the desired derivatives **9h**−**j** as for allyl derivative **9g**.

Inhibition Assay Results. Enol mimics **5** and **9** were assayed in the presence of 3-dehydroquinic acid (**1**) for their inhibitory properties of DHQ2-Mt. All of the compounds proved to be competitive reversible inhibitors of the enzyme. The inhibition data are summarized in Table 1.

In general, the monosubstituted compounds **5** proved to be more potent than the disubstituted series **9** (Table 1, entries 1−6 vs 7−12). Among compounds **5**, significant differences were not found between the different aromatic moieties, although the presence of a larger ring provides slightly more active compounds (Table 1, entries 1−5 vs 6). With the exception of thiophene **5f**, compounds **5** have inhibition constants in the low nanomolar range (28−43 nM), with benzothiophene **5a** being the most potent in the series with a K_i of 28 nM.

In general, substitution of C2 in **5** leads to a loss of activity. However, with some exceptions, the disubstituted compounds **9** still have inhibition constants in the nanomolar range. Although the disubstituted compounds **9** do not follow an absolutely clear pattern, certain trends can be identified. In general, the

dx.doi.org/10.1021/jm2006063 |J. Med. Chem. 2011, 54, 6063–6084

Table 2. Crystallographic Data Collection and Refinement Statistics for the DHQ2-Mt Complex with Inhibitors 5b, 9d, and 9h

data processing[a]	DHQ2-Mt/**5b**	DHQ2-Mt/**9d**	DHQ2-Mt/**9h**
space group	*F*23	*F*23	*F*23
cell parameters (Å)[b]	$a = b = c = 126.63$	$a = b = c = 126.43$	$a = b = c = 125.80$
wavelength (Å)	0.9724	0.8726	0.9921
detector	ADSC Q315r CCD	MarMosaic225 CCD	ADSC Q210r CCD
observed reflections[c]	26866 (3925)[d]	5918 (858)[d]	30571 (3015)[d]
resolution range (Å)	29.06−1.50 (1.58−1.50)	38.00−2.50 (2.64−2.50)	31.45−1.40 (1.48−1.40)
Wilson B ($Å^2$)	17.0	28.6	16.8
multiplicity	7.8 (4.0)	14.5 (14.6)	7.5 (1.6)
completeness	0.995 (0.998)	1.000 (1.000)	0.943 (0.649)
R_{merge}	0.062 (0.367)	0.120 (0.388)	0.062 (0.397)
	Refinement[e]		
resolution range (Å)	21.41−1.50 (1.58−1.50)	20.00−2.50 (2.63−2.50)	20.00−1.50 (1.58−1.50)
reflections used in refinement[d]	25423 (3691)	5397 (775)	24982 (3601)
reflections used for R_{free}	1443 (223)	508 (83)	1330 (192)
R factor[f]	0.127 (0.174)	0.162 (0.173)	0.162 (0.221)
R_{free}[g]	0.161 (0.215)	0.211 (0.220)	0.184 (0.262)
rmsd (bonds (Å)/angles (deg))	0.014/1.5	0.012/1.5	0.014/1.5
	Final Model		
protein/inhibitor/sodium/sulfate/water atoms	1091/24/2/25/1/129	1039/43/-/15/52	1079/46/-/20/107
average B protein ($Å^2$)/inhibitor ($Å^2$)/ sulfate ($Å^2$)/sodium ($Å^2$)/water ($Å^2$)	16.7/21.4/34.3/40.7/36.3	14.5/31.2/-/36.21/15.2	14.0/16.6/-/21.4/28.5
Ramachandran statistics[h] (%)	98.5/100.0	98.5/100.0	98.5/100.0
PDB accession code	2Y71	2Y76	2Y77

[a] Results from SCALA.[45] [b] One Ångstrom (Å) is 0.1 nm. [c] No σ cutoff or other restrictions were used for inclusion of reflections. [d] Values in parentheses are for the highest resolution bin, where applicable. [e] Results from REFMAC.[44] [f] *R*-factor = $\Sigma||F_{obs}(hkl)| - |F_{calc}(hkl)|| / \Sigma|F_{obs}(hkl)|$. [g] According to Brunger.[42] [h] According to the program MOLPROBITY.[47] The percentages indicated are for residues in favored and total allowed regions, respectively.

substitution of position 2 with an allyl group reduces the binding affinity vs a benzyl group (Table 1, entries 7 and 14 vs 13). For compounds **9a**−**f**, which are substituted with the same aromatic ring in both positions 2 and 3, the 2-benzothiophene moiety gives higher binding affinities to the *M. tuberculosis* enzyme. Benzothiophene **9a** proved to be the most potent compound in the series with a K_i of 40 nM. In the case of compounds **9h**−**j**, which are susbstituted with different aromatic rings in positions 2 and 3, compound **9j** proved to be the most potent one with a K_i of 59 nM. The inhibition results for the disubstituted compounds **9** suggest that, in addition to the size and the type of aromatic ring, the relative disposition of the two substituents in the active site might also affect the binding affinity. Structural and molecular modeling studies on these compounds were therefore required and will be discussed in the next section.

Compounds **5** and **9** also proved to be competitive reversible inhibitors of the *H. pylori* enzyme but with lower K_i values (Table 1). Among the monosubstituted compounds **5**, benzothiophenes **5a** and **5b** proved to be the most potent inhibitors, with inhibition constants of 132 and 130 nM, respectively. As far as the *M. tuberculosis* enzyme is concerned, among compounds **9a**−**f**, which are substituted with the same aromatic ring in both positions 2 and 3, compounds **9a** and **9b** proved to be the most potent, with inhibition constants of 97 and 50 nM, respectively. With the exception of compound **9c**, the affinity remained almost unchanged when a benzyl substituent was present on C2 (Table 1, entries 7−8 and 10−11 vs 1−2 and 4−5). For compounds **9h**−**j**, with different aromatic rings in positions 2 and 3, the presence of an aromatic moiety on C2 is clearly more favorable than an allyl group (i.e., compound **9g**). Compound **9j** also proved to be the most potent of this series, with a K_i of 100 nM.

Structural Studies. To obtain structural information on the binding mechanism of these inhibitors, crystal structures of the methylbenzothiophene **5b** and disubstituted compounds **9d** and **9h** in complex with DHQ2-Mt were solved at 1.5, 2.5, and 1.5 Å, respectively. DHQ2-Mt/**9d** and DHQ2-Mt/**9h** binary complexes were obtained by cocrystallization, and the DHQ2-Mt/**5b** binary complex crystals were obtained by soaking apo-DHQ2-Mt crystals. Data were collected from cryocooled crystals using synchrotron radiation and were processed (Table 2). The structures were solved by molecular replacement, using the crystal structure of DHQ2-Mt bound to an oxime solved by Lapthorn et al. (PDB entry 1H0S[33]) as a search model and then refined (Figure 4). All three complexes contain a single DHQ2-Mt molecule in the crystallographic asymmetric unit.

Comparison of DHQ2-Mt/**5b** and our recently solved crystal structure of DHQ2-Mt in complex with (2*R*)-2-(4-methoxy)benzyl-3-dehydroquinic acid (PDB entry 2XB8[17]) shows that both structures are virtually identical (0.11 Å root-mean-square difference after superposition of 121 α-carbon atoms) with the exception of three amino acids located on the loop, residues 18−20, which are not visible in the structure reported here (Figure 5a). In PDB entry 2XB8, amino acids belonging to the loop (residues 18−25) also showed more disorder than the rest of the protein. The cyclohexene core of **5b** occupies approximately the same site as the cyclohexane ring of (2*R*)-2-(4-methoxy)benzyl-3-dehydroquinic acid in PDB entry 2XB8.

dx.doi.org/10.1021/jm2006063 |*J. Med. Chem.* 2011, 54, 6063–6084

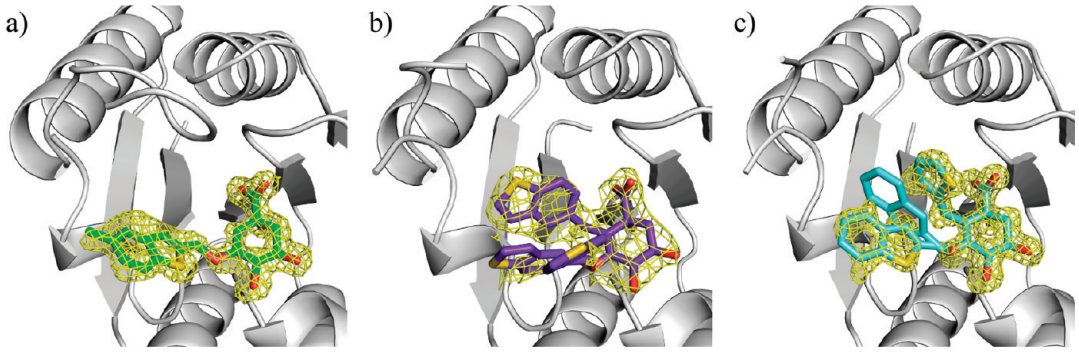


Figure 4. Unbiased electron density for inhibitors: (a) **5b**; (b) **9d**; (c) **9h**. From the model obtained by molecular replacement, refinement was performed to obtain unbiased density for the inhibitor molecule and other model changes. A maximum-likelihood weighted $2F_o - F_c$ map[44] contoured at 1σ is shown up to 1.6 Å around the inhibitor molecule (yellow). The final model (gray), including the inhibitor molecule [**5b** (green); **9d** (purple); **9h** (cyan)], is superposed onto the map. Inhibitors **9d** and **9h** both showed alternative conformations for their *O*-alkyl substituents.

Calculated maps showed clear high electron density for the inhibitor molecule **5b**. The benzothiophene ring of **5b** is located with its sulfur atom orientated at the same side as the oxygen of C4, as shown in Figure 4a. The higher electron density obtained at this side of the aromatic ring, as well as in its *para* position, suggests that the orientation of the benzothiophene ring is as indicated in Figure 4a and not the other putative rotamer around the methylene group. This binding mode may be due to two factors; first, in this disposition the methyl group can interact with the carbon side chain of Arg15 and, second, in the other putative conformation the methyl group would be located too close to Glu92 of a symmetry-related neighboring molecule.

Inhibitor **5b** fits snugly in the active site by establishing a large number of polar interactions (carboxylate and hydroxyl groups), including the essential water molecule involved in the enzymatic reaction (Figure 5b). This crystal structure also shows that the aromatic moiety of inhibitor **5b** interacts with the apolar part of the active site by establishing important lipophilic interactions, i.e., it is in close contact with the side chain carbons of Arg15 and Leu13 and the main chain of Leu13 and Leu16 (and with side chain carbon atoms of Glu92 of the symmetry-related molecule). The methylene group of the inhibitor side chain is in close contact with the side chain of Leu13 (Figure 5c). Moreover, the five-membered ring of the benzothiophene moiety interacts with the essential Tyr24 through a CH$-\pi$ interaction.

As observed for PDB entry 2XB8 and the crystal structure of methylbenzothiophene **5b** in complex with DHQ2-Hp (PDB entry 2WKS[28]), the essential arginine of the loop, Arg19, is probably located outside the active site, with its position occupied by the aromatic moiety of the inhibitor. Molecular dynamics calculations suggest that the required orientation of Tyr24 for proton abstraction is induced by the proximity of Arg19.[17] Therefore, the conformational changes in the loop that predominantly affect the inappropriate orientation of the catalytic tyrosine and the inability of the essential arginine to be located close to it are probably responsible for the high inhibition potency of methylbenzothiophene **5b**. Similar behavior is expected for the monosubstituted inhibitors **5a**−**e**.

There is no significant difference between the protein structures in the DHQ2-Mt/**9d** and the DHQ2-Mt/**9h** binary complexes (root-mean-square difference of 0.12 Å after superposition of 128 α-carbon atoms). Therefore, an explanation for the different crystallographic resolutions obtained is not obvious (2.5 and 1.5 Å, respectively) (Figure 6). We can only hypothesize that the larger substituent on C2 (the benzo[*b*]thiophen-5-ylmethoxy moiety of **9d** vs the thien-2-ylmethyl moiety of **9h**) and/or the different C3 substituent (the benzo[*b*]thiophen-5-ylmethoxy moiety of **9d** vs the benzo[*b*]thiophen-2-ylmethoxy moiety of **9h**) somehow lead to more disorder of the covering loop and perhaps to disturbance of certain crystal contacts. In both binary complexes, the benzyl moiety on C2 is located approximately perpendicular to the position that should be occupied by the side chain of the catalytic Tyr24. The substituent on C2 seems to block the approach of the catalytic tyrosine to Arg108, thus preventing the formation of the hydrogen-bonding interaction required to close the active site upon substrate binding. The proximity of both residues, together with the essential Arg19, is responsible for lowering the pK_a of Tyr24. In addition, as for methylbenzothiophene **5b**, the substituent on C3 should block the entrance of the catalytic arginine into the active site. Therefore, the disubstituted inhibitors appear to inhibit the enzyme by a double effect. On the one hand, they avoid the closure of the active site by preventing proper alignment of Arg108 and Tyr24 and, on the other hand, they block the entry of Arg19 into the active site. The latter residue appropriately orients the catalytic tyrosine to initiate the enzymatic reaction and is also responsible for the basicity of the tyrosine.

In Vitro Activity: Synthesis of Ester Prodrugs. The in vitro antibacterial activity of our inhibitors was studied by determining the minimum inhibitory concentration (MIC) against *M. tuberculosis* H37Rv by using the Alamar Blue Assay.[34] In general, activity was not observed for compounds **5** and **9** below 200 μg/mL. We assume that this lack of activity could be due to the high hydrophilicity of these compounds. It is known that hydrophilic agents cross the mycobacterial cell wall slowly because the mycobacterial porin is inefficient in allowing the permeation of solutes and is only present in low amounts.[35] To improve permeability and therefore antibacterial activity, lipophilic prodrugs **6**−**8** and **10**−**11** were designed. We hypothesized that these compounds would be slowly hydrolyzed to the carboxylate active form after absorption by the bacterium. Three types of esters with different stabilities against hydrolysis were synthesized and evaluated. In addition, transformation of secondary hydroxyl groups into butyryl esters was also studied.

First, ester prodrugs of inhibitors **5**, compounds **6**−**8**, were synthesized. Yields are shown in Table 3. Methyl esters **6** were prepared from the corresponding lactones **15** by treatment with sodium methoxide in methanol. Ethyl derivatives **7** and propyl

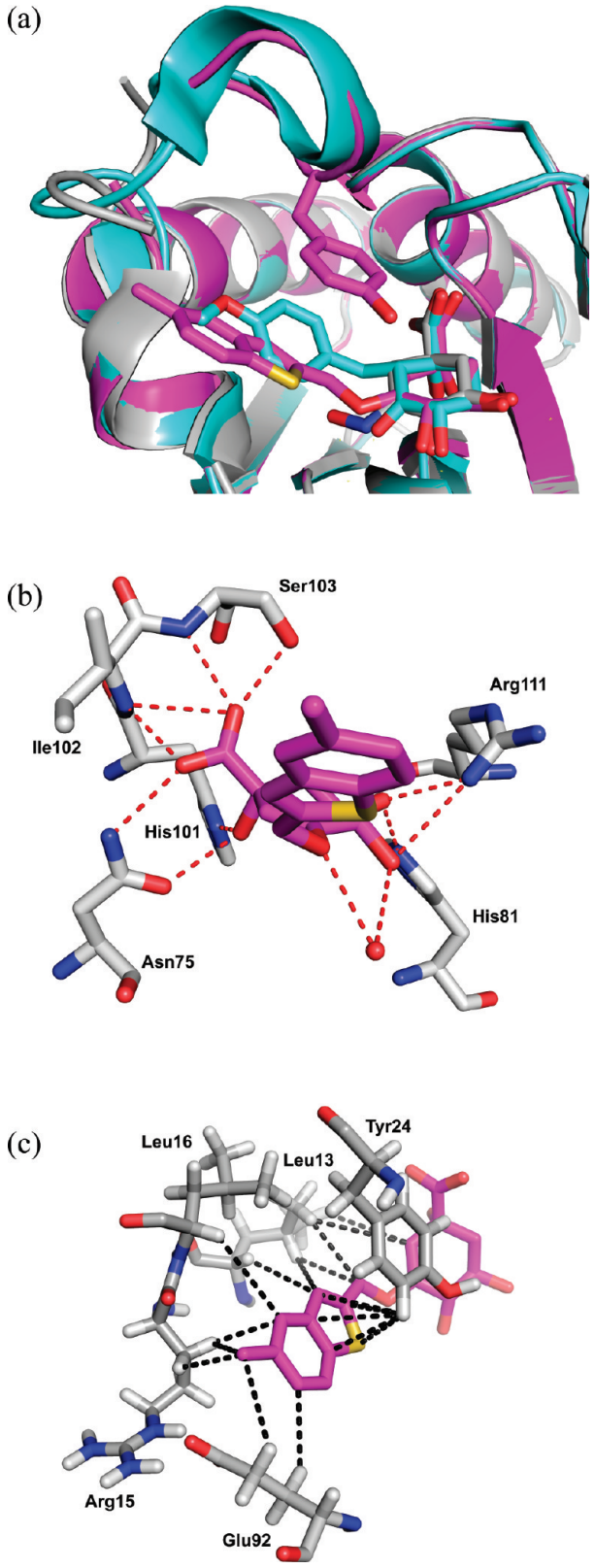


Figure 5. (a) Superposition of DHQ2-Mt structure in complex with oxime (PDB entry 1H0S,[41] gray), (2*R*)-2-(4-methoxy)benzyl-3-dehydroquinic acid (PDB entry 2XB8,[17] cyan), and **5b** (PDB entry 2Y71, magenta). Residues 18−20 are not solved in 2Y71. (b) Polar contacts (red dashes) between ligand **5b** and DHQ2-Mt. (c) Apolar contacts (black dashes) between ligand **5b** and DHQ2-Mt. Only contacts less than 3.3 Å are indicated.

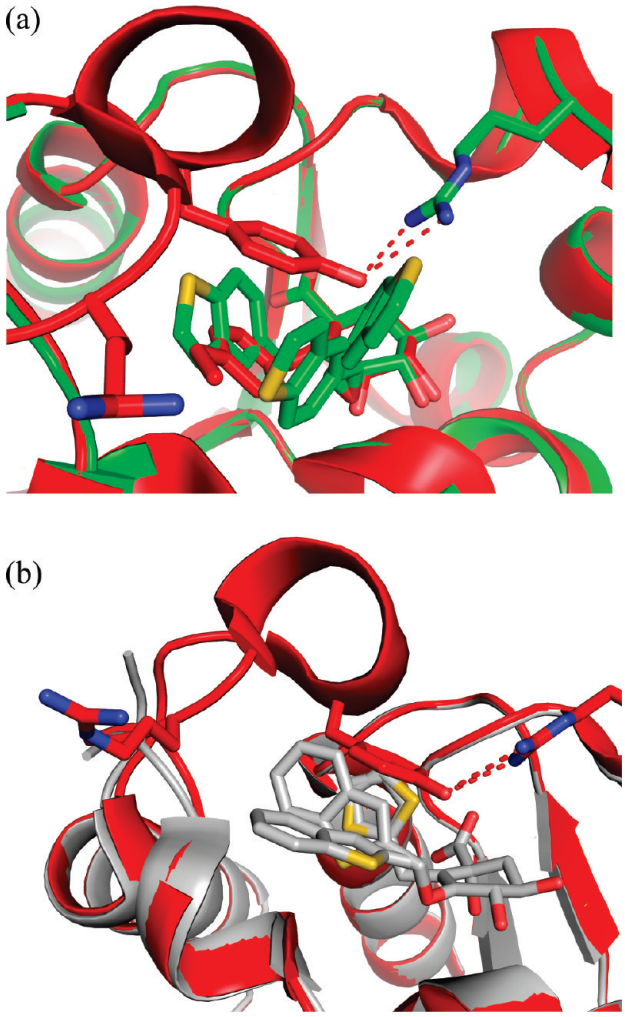
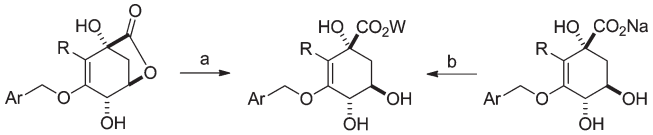


Figure 6. Superposition of DHQ2-Mt structure in complex with (2*R*)-2-(4-methoxy)benzyl-3-dehydroquinic acid (PDB entry 2XB8,[17] red) with: (a) DHQ2-Mt/**9d** binary complex (PDB entry 2Y76, green); (b) DHQ2-Mt/**9h** binary complex (PDB entry 2Y77, gray). The loop is disordered in PDB entries 2Y76 and 2Y77.

esters **8** were synthesized by a nucleophilic substitution of ethyl bromide and propyl bromide by sodium salts **5**, respectively. A similar strategy was employed for the synthesis of disubstituted compounds **10** and **11** (Table 3).

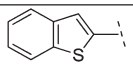
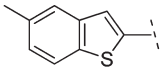
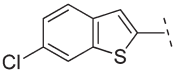
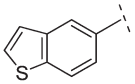
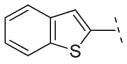
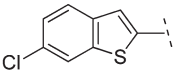
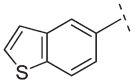
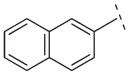
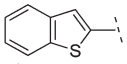
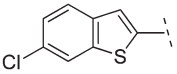
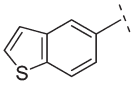
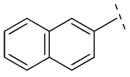
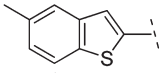
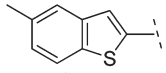
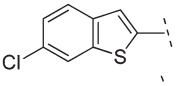
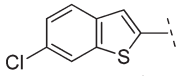
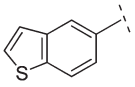
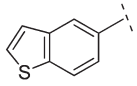
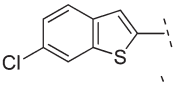
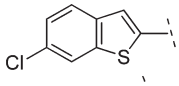
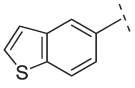
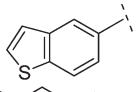
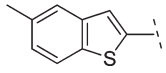
The antibacterial activity of compounds **6**−**8** and **10**−**11** against *M. tuberculosis* H37Rv was determined, and the results are summarized in Table 4. In general, the activity dramatically increases with the stability of the ester, i.e., propyl esters **8** are more active than the corresponding methyl esters **6** (Table 4, entries 1 and 4 vs 12 and 14). The propyl esters **8a**, **8f**, and **8g** proved to be the most potent of the series with an MIC value of 5 μg/mL (Scheme 4). Furthermore, ethyl esters showed higher MIC values than the corresponding propyl esters. However, the in vitro activity of ethyl esters **7** can be increased by esterification of their secondary hydroxyl groups as butyryl esters (Table 4, entry 7 vs 9). Thus, ethyl esters **7e** and **7f** gave an MIC value of 5 μg/mL. It is important to highlight that esterification of all free hydroxyl groups provides MIC values that are as high as those of the methyl ester derivatives **6** (>160 μg/mL). In addition, esterification to give less stable esters, such as acetyl esters, is also less efficient.

Table 3. Synthesis of Esters 6−8 and 10−11



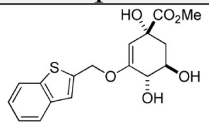
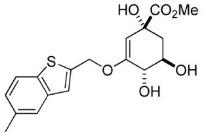
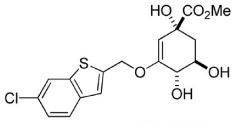
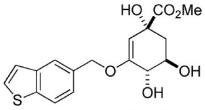
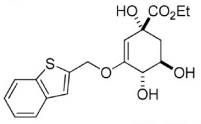
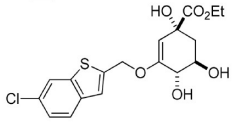
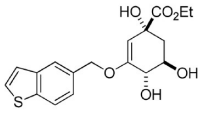
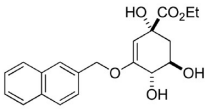
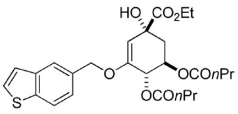
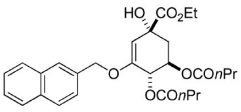
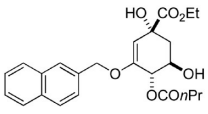
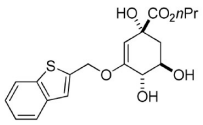
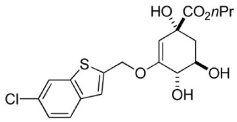
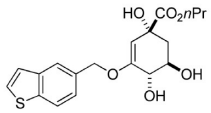
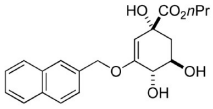
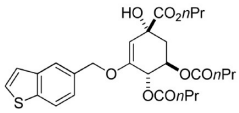
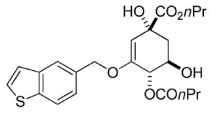
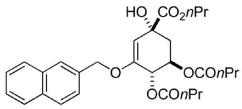
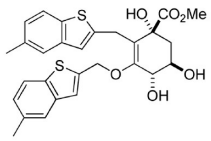
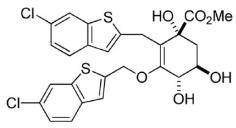
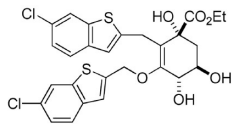
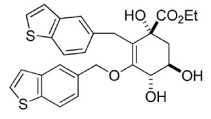
15, 16 **6–8, 10–11** **5, 9**

(a) MeONa, MeOH, RT; (b) EtBr or *n*PrBr, DMF, RT.

Starting material	Product	Ar	R	W	Yield (%)
15a	**6a**		H	Me	53
15b	**6b**		H	Me	62
15c	**6c**		H	Me	46
15d	**6d**		H	Me	68
5a	**7a**		H	Et	49
5c	**7b**		H	Et	46
5d	**7c**		H	Et	58
5e	**7d**		H	Et	42
5a	**8a**		H	*n*Pr	38
5c	**8b**		H	*n*Pr	38
5d	**8c**		H	*n*Pr	32
5e	**8d**		H	*n*Pr	50
16b	**10a**			Me	24
16c	**10b**			Me	49
16d	**10c**			Me	40
16f	**10d**			Me	51
9c	**11a**			Et	21
9d	**11b**			Et	49
9i	**11c**			Et	37

dx.doi.org/10.1021/jm2006063 |*J. Med. Chem.* 2011, 54, 6063–6084

Table 4. The Minimum Inhibitory Concentration (MIC, μg/mL) of Esters 6−11 against *M. tuberculosis* H37Rv

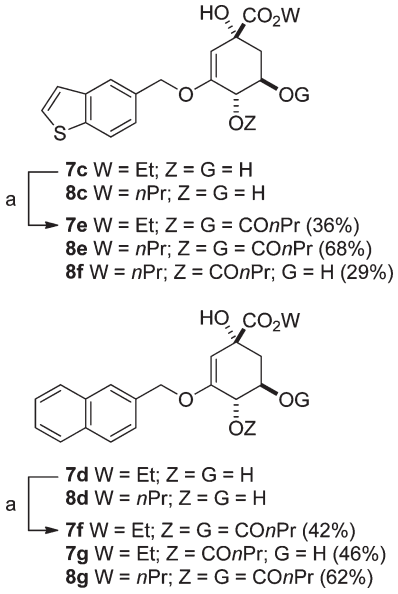
Entry	Compound	MIC
1	 **6a**	>160
2	 **6b**	>160
3	 **6c**	>160
4	 **6d**	>160
5	 **7a**	40
6	 **7b**	40
7	 **7c**	40
8	 **7d**	20
9	 **7e**	5
10	 **7f**	5
11	 **7g**	10
12	 **8a**	5
13	 **8b**	40
14	 **8c**	20
15	 **8d**	10
16	 **8e**	20
17	 **8f**	5
18	 **8g**	5
19	 **10a**	>160
20	 **10b**	80
21	 **11a**	>160
22	 **11b**	160

Furthermore, 2,3-disubstituted ester derivatives **10**−**11** are less active than the corresponding monosubstituted compounds **6**−**7**. This finding could be due to the higher volume of compounds **10**−**11**, a characteristic that hinders their entry into the bacterium.

The cytotoxicity of esters **6**−**11** was evaluated against human fibroblast cells (MRC-5) using the crystal violet toxicity assay protocol. No toxicity of cell growth was observed up to the highest concentration tested, 300−400 μg/mL.

dx.doi.org/10.1021/jm2006063 |*J. Med. Chem.* 2011, 54, 6063–6084

Scheme 4. Synthesis of Ethyl Esters 7e−g and Propyl Esters 8e−g[a]



[a] Reagents and conditions: (a) nPrCOCl, Py, CH_3CN, RT.

CONCLUSIONS AND FINAL REMARKS

Several enol mimics, compounds **5** and **9**, of the reaction catalyzed by DHQ2, the third enzyme of the shikimic acid pathway, have been synthesized and tested. All compounds proved to be reversible competitive inhibitors of DHQ2 from *M. tuberculosis* and *H. pylori*, which are essential enzymes for these pathogenic bacteria. The effect of the substituent at C2 of enol mimics **5** was studied with the 2,3-disubstituted compounds **9**. The studies reported here showed that the introduction of a benzyl group on C2 led to a decrease in the inhibition potency, but in most cases the inhibition constants are also in the nanomolar range. Benzothiophene derivative **5a** proved to be the most potent of the monosubstituted series with a K_i of 28 nM against DHQ2-Mt. The corresponding disubstituted derivative **9a** showed a K_i of 40 nM. The substitution of both C2 and C3 with different benzyl moieties also provided good inhibition potencies, with compound **9j** having a K_i value of 59 nM.

Compounds **5** and **9** also proved to be good competitive inhibitors of DHQ2-Hp. The disubstituted derivatives **9a** and **9b** were the most potent compounds, with inhibition constants of 97 and 50 nM, respectively.

Monosubstituted benzothiophene derivative **5b** and double substituted compounds **9d** and **9h** were cocrystallized with DHQ2-Mt, and their structures were solved at 1.5, 2.5, and 1.5 Å, respectively. Compound **5b** binds to DHQ2-Mt in a similar way to the previously reported DHQ2-Hp/**5b** complex (PDB entry 2WKS[28]). However, in the binary DHQ2-Mt/**5b** complex reported here, the benzothiophene ring is rotated by 180° to orient the methyl group of the aromatic ring in such a way that it is in close contact with the carbon side chain of Arg15. The aromatic moiety of inhibitor **5b** interacts with the apolar part of the active site (side chain of Arg15, Leu13, Leu16, etc.) and expels the essential arginine of the loop from the active site. It has been suggested that the reaction is initiated by the proximity of this essential arginine, which appropriately orients the essential tyrosine to initiate the enzymatic reaction.[16,17]

The crystal structures of binary complexes DHQ2-Mt/**9d** and DHQ2-Mt/**9h** suggest that these 2,3-disubstituted compounds might inhibit the enzyme by a double effect. Thus, these inhibitors avoid the closure of the active site by preventing hydrogen-bonding interaction between the essential Tyr24 and Arg108 and they also block the entry of Arg19 in the active site. Therefore, compounds that are able to block the alignment between Tyr24 and Arg108 would also be good competitive inhibitors of DHQ2 enzymes.

An ester prodrug approach was used to improve permeability of the mycobacterial cell. These studies showed that the stability of the ester is crucial to achieve improved in vitro activities. Propyl esters **8** were the most efficient in achieving improved in vitro activities, which are in the low micromolar range. Ethyl esters **7** can also be used, but esterification of the free secondary hydroxyl groups as butyryl esters is required. In this way MIC values similar to those of the propyl esters **8** could be achieved. The results obtained highlight the possibility that inhibitors of the shikimic acid pathway could be used as new anti-TB agents and reveal the importance of improving inhibitor hydrophilicity to traverse the mycobacteria wall.

EXPERIMENTAL SECTION

General Procedures. All starting materials and reagents were commercially available and were used without further purification. ^{1}H NMR spectra (250, 300, 400, and 500 MHz) and ^{13}C NMR spectra (63, 75, 100, and 125 MHz) were measured in deuterated solvents. *J* values are given in hertz. NMR assignments were carried out by a combination of 1 D, COSY, and DEPT-135 experiments. FT-IR spectra were recorded as NaCl plates or KBr discs. $[\alpha]_D^{20}$ values are given in 10^{-1} deg cm^2 g^{-1}. Purity of compounds **5**−**11** were determined by a combination of ^{1}H NMR and reverse-phase and normal-phase HPLC and were found to be >95%. Compounds **5a**−**b**, **5d**, **5f**, **14a**−**b**, **14d**, and **14f** were prepared as described previously.[28]

2-Bromomethyl-6-chlorobenzo[*b*]thiophene. A stirred solution of 6-chlorobenzo[*b*]thiophen-2-ylmethanol (400 mg, 2.01 mmol) and PPh_3 (950 mg, 3.62 mmol) in dry dichloromethane (150 mL) was treated with CBr_4 (801 mg, 2.42 mmol) under argon. After stirring for 1 h, diethyl ether was added and the resulting precipitate was filtered off and washed with diethyl ether. The filtrate was concentrated under reduced pressure, and the resulting residue was purified by flash chromatography eluting with (30:70) diethyl ether/hexanes to give 2-bromomethyl-6-chlorobenzo[*b*]thiophene (483 mg, 92%) as a white amorphous solid; mp 79−81 °C. ^{1}H NMR (250 MHz, $CDCl_3$) δ 7.74 (d, *J* = 1.5 Hz, 1H, H-7), 7.59 (d, *J* = 8.7 Hz, 1H, H-4), 7.30 (dd, *J* = 8.7 and 1.5 Hz, 1H, H-6), 7.24 (s, 1H, H-3) and 4.75 (s, 2H, CH_2) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ 141.6 (C), 141.4 (C), 137.5 (C), 130.9 (C), 125.4 (CH), 124.5 (CH), 123.8 (CH), 121.9 (CH), and 26.8 (CH_2) ppm. MS (EI) *m*/*z* (%) 260 and 262 (M^+). HRMS calcd for $C_9H_6ClSBr^{79}$ (M^+): 259.9062; found, 259.9067.

(1*R*,4*S*,5*R*)-1,4-Di(*tert*-butyldimethylsilyloxy)-3-(6-chlorobenzo[*b*]thiophen-5-yl)methoxycyclohex-2-en-1,5-carbolactone (13c) and (1*R*,4*S*,5*R*)-1,4-Di(*tert*-butyldimethylsilyloxy)-3-(6-chlorobenzo[*b*]thiophen-5-yl)methoxy-2-(6-chlorobenzo[*b*]-thiophen-5-yl)methylcyclohex-2-en-1,5-carbolactone (14c). A flame-dried round-bottomed flask was charged with KHMDS (2.32 mL, 1.16 mmol, 0.5 M in toluene) and dry DMF (1.16 mL) under an inert atmosphere. The resultant solution was cooled to −78 °C, and a solution of ketone **12**[22] (232 mg, 0.58 mmol) in a 1:1 mixture of DMF and toluene (5.8 mL), both dry, was added. The mixture was stirred for 30 min, and a solution of 2-bromomethyl-6-chlorobenzo[*b*]thiophene (300 mg, 1.42 mmol) in a 3:2 mixture of DMF and toluene (4.8 mL),

both dry, was added. After 45 min, the reaction mixture was diluted successively with diethyl ether and water. The organic phase was separated and the aqueous layer was extracted with diethyl ether (×2). The combined organic extracts were dried (anhyd Na_2SO_4), filtered, and evaporated under reduced pressure. The residue was purified by flash chromatography eluting with diethyl ether/hexanes [1) 0:100, 2) 5:95] to afford *O*-alkyl derivative **13c** (47 mg, 23%) and dialkyl derivative **14c** (142 mg, 46%). Data for **14c**: beige solid; mp 153−155 °C. $[\alpha]_D^{20}$ = −116° (*c* 1.2, $CHCl_3$). ^{1}H NMR (250 MHz, $CDCl_3$) δ 7.55 (d, *J* = 1.7 Hz, 1H, ArH), 7.50 (d, *J* = 1.7 Hz, 1H, ArH), 7.38 (d, *J* = 8.5 Hz, 1H, ArH), 7.26 (d, *J* = 8.5 Hz, 1H, ArH), 7.07 (m, 2H, 2 × ArH), 6.83 (s, 1H, ArH), 6.72 (s, 1H, ArH), 4.91 (d, *J* = 12.5 Hz, 1H, OC*H*H), 4.84 (d, *J* = 12.5 Hz, 1H, OCH*H*), 4.44 (dd, *J* = 5.7 and 3.5 Hz, 1H, H-5), 4.33 (d, *J* = 3.5 Hz, 1H, H-4), 3.75 (d, *J* = 15.7 Hz, 1H, C*H*HAr), 3.62 (d, *J* = 15.7 Hz, 1H, CH*H*Ar), 2.41 (d, *J* = 11.0 Hz, 1H, H-6_{eq}), 2.30 (dd, *J* = 11.0 and 5.7 Hz, 1H, H-6_{ax}), 0.81 (s, 9H, $C(CH_3)_3$), 0.62 (s, 9H, $C(CH_3)_3$), 0.06 (s, 3H, CH_3), 0.05 (s, 3H, CH_3), 0.01 (s, 3H, CH_3) and −0.08 (s, 3H, CH_3) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ 175.1 (C), 148.5 (C), 144.6 (C), 141.0 (C), 140.2 (2 × C), 138.4 (C), 137.5 (C), 130.5 (C), 129.0 (C), 128.6 (CH), 125.1 (CH), 124.5 (C), 124.3 (CH), 123.3 (CH), 121.8 (CH), 121.4 (CH), 120.7 (CH), 74.6 (C), 74.5 (CH), 67.9 (CH_2), 67.2 (CH), 37.4 (CH_2), 25.7 ($C(CH_3)_3$), 25.4 (CH_2 + $C(CH_3)_3$), 18.0 ($C(CH_3)_3$), 18.0 ($C(CH_3)_3$), −3.3 (CH_3), −3.5 (CH_3), −4.4 (CH_3) and −4.5 (CH_3) ppm. IR (KBr) 1792 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 761 (MH^+). HRMS calculated for $C_{37}H_{47}O_5S_2Si_2Cl_2$ (MH^+), 761.1775; found, 761.1754.

(1*R*,4*S*,5*R*)-1,4-Dihydroxy-3-(6-chlorobenzo[*b*]thiophen-5-yl)methoxycyclohex-2-en-1,5-carbolactone (15c). A stirred solution of silyl ether **13c** (164 mg, 0.28 mmol) in dry THF (4.0 mL) was treated with tetrabutylammonium fluoride (0.62 mL, 0.62 mmol, ca. 1.0 M in THF). After 1 h, ethyl acetate and water were added. The organic layer was separated, and the aqueous phase was extracted with ethyl acetate (×3). The combined organic extracts were dried (anhyd Na_2SO_4), filtered, and concentrated under reduced pressure. The resulting residue was purified by flash chromatography eluting with ethyl acetate/hexane (50:50) to give diol **15c** (31 mg, 31%) as a white foam; mp 79−81 °C. $[\alpha]_D^{20}$ = −116° (*c* 1.6, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.88 (d, *J* = 1.7 Hz, 1H, ArH), 7.73 (d, *J* = 8.5 Hz, 1H, ArH), 7.37 (d, *J* = 0.7 Hz, 1H, ArH), 7.33 (dd, *J* = 8.5 and 1.7 Hz, 1H, ArH), 5.19 (s, 1H, H-2), 5.12 (d, *J* = 13.0 Hz, 1H, OC*H*H), 5.06 (d, *J* = 13.0 Hz, 1H, OCH*H*), 4.63 (m, 1H, H-5), 4.12 (d, *J* = 3.5 Hz, 1H, H-4) and 2.32 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 179.1 (C), 155.3 (C), 142.8 (C), 142.0 (C), 139.4 (C), 131.6 (C), 126.2 (CH), 125.8 (CH), 124.1 (CH), 122.9 (CH), 105.6 (CH), 77.0 (CH), 73.0 (C), 67.7 (CH), 66.2 (CH_2) and 38.3 (CH_2) ppm. IR (KBr) 3398 (O−H) and 1770 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 375 (MNa^+). HRMS calcd for $C_{16}H_{13}O_5SClNa$ (MNa^+), 375.0064; found, 375.0062.

Sodium (1*R*,4*S*,5*R*)-3-(6-Chlorobenzo[*b*]thiophen-5-yl)-methoxy-1,4,5-trihydroxycyclohex-2-ene-1-carboxylate (5c). A solution of lactone **15c** (8 mg, 0.02 mmol) in THF (0.2 mL) and aqueous sodium hydroxide (45 μL, 0.02 mmol, 0.5M) was stirred at room temperature for 15 min. Water was added, and THF was removed under reduced pressure. The resulting aqueous solution was washed with diethyl ether (×2), and the aqueous extract was lyophilized to afford derivative **5c** (9 mg, 99%) as a white solid; mp 200−202 °C (dec). $[\alpha]_D^{20}$ = −10° (*c* 0.5, H_2O). ^{1}H NMR (400 MHz, D_2O/CD_3CN (2:1)) δ 8.40 (d, *J* = 2.0 Hz, 1H, ArH), 8.26 (d, *J* = 8.4 Hz, 1H, ArH), 7.86 (s, 1H, ArH), 7.85 (dd, *J* = 8.4 and 2.0 Hz, 1H, ArH), 5.54 (d, *J* = 12.4 Hz, 1H, OC*H*H), 5.47 (d, *J* = 12.4 Hz, 1H, OCH*H*), 5.34 (s, 1H, H-2), 4.43 (d, *J* = 6.0 Hz, 1H, H-4), 4.35 (m, 1H, H-5) and 2.47 (m, 2H, CH_2-6) ppm. ^{13}C NMR (100 MHz, D_2O/CD_3CN (2:1)) δ 180.9 (C), 155.6 (C), 141.3 (C), 138.3 (C), 130.2 (C), 125.4 (CH), 125.3 (CH), 123.3 (CH), 122.2 (CH + C), 102.0 (CH), 73.7 (C), 71.4 (CH), 69.9 (CH), 65.0 (CH_2) and 37.2 (CH_2) ppm. IR (KBr) 3436 (O−H) and 1683 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 369 (M − H). HRMS calculated for $C_{16}H_{13}O_6SClNa$ (M − H), 391.0014; found, 391.0027.

(1*R*,4*S*,5*R*)-2-(6-Chlorobenzo[*b*]thiophen-5-yl)methyl-3-(6-chlorobenzo[*b*]thiophen-5-yl)methoxy-1,4-dihydroxy-cyclohex-2-en-1,5-carbolactone (16c). The experimental procedure was the same as for compound **15c** using silyl ether **14c** (144 mg), TBAF (0.42 mL), and THF (2.7 mL). Yield = 67 mg (66%). Yellow solid; mp 201−202 °C. $[\alpha]_D^{20}$ = −152° (*c* 0.6, MeOH/acetone (2:1)). ^{1}H NMR (250 MHz, DMSO-d_6) δ 8.08 (d, *J* = 2.0 Hz, 1H, ArH), 7.93 (d, *J* = 2.0 Hz, 1H, ArH), 7.79 (d, *J* = 8.5 Hz, 1H, ArH), 7.61 (d, *J* = 8.5 Hz, 1H, ArH), 7.39 (dd, *J* = 8.5 and 2.0 Hz, 1H, ArH), 7.39 (s, 1H, ArH), 7.28 (dd, *J* = 8.5 and 2.0 Hz, 1H, ArH), 7.04 (s, 1H, ArH), 6.39 (s, 1H, OH), 6.12 (d, *J* = 7.0 Hz, 1H, OH), 5.37 (d, *J* = 13.0 Hz, 1H, OC*H*H), 5.28 (d, *J* = 13.0 Hz, 1H, OCH*H*), 4.62 (m, 1H, H-5), 4.49 (dd, *J* = 7.0 and 3.5 Hz, 1H, H-4), 3.81 (d, *J* = 14.7 Hz, 1H, C*H*HAr), 3.67 (d, *J* = 14.7 Hz, 1H, CH*H*Ar) and 2.29 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, DMSO-d_6) δ 176.2 (C), 147.8 (C), 145.5 (C), 142.1 (C), 140.6 (C), 140.2 (C), 138.4 (C), 137.7 (C), 129.2 (C), 127.9 (C), 125.0 (CH), 124.9 (CH), 124.3 (CH), 123.8 (CH), 122.3 (CH), 122.0 (CH), 121.5 (CH), 121.3 (C), 120.8 (CH), 74.6 (CH), 71.7 (C), 64.8 (CH_2), 64.3 (CH), 36.8 (CH_2) and 24.5 (CH_2) ppm. IR (KBr) 3408 (O−H) and 1749 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 555 (MNa^+). HRMS calcd for $C_{25}H_{18}O_5S_2Cl_2Na$ (MNa^+), 554.9865; found, 554.9865.

Sodium (1*R*,4*S*,5*R*)-3-(6-Chlorobenzo[*b*]thiophen-5-yl)-methoxy-2-(6-chlorobenzo[*b*]thiophen-5-yl)methyl-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (9c). The experimental procedure was the same as for compound **5c** using lactone **16c** (8 mg), NaOH (30 μL) and THF (0.1 mL). Yield = 8 mg (93%). Beige solid; mp 189 °C (dec). $[\alpha]_D^{20}$ = −42° (*c* 0.8, (2:1) DMSO/MeOH). ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.65 (d, *J* = 8.0 Hz, 1H, ArH), 7.71 (d, *J* = 9.0 Hz, 1H, ArH), 7.53 (d, *J* = 8.5 Hz, 1H, ArH), 7.35 (dd, *J* = 8.5 and 2.0 Hz, 1H, ArH), 7.23 (m, 2H, 2 × ArH), 7.02 (s, 1H, ArH), 5.21 (s, 1H, OH), 5.16 (d, *J* = 13.0 Hz, 1H, C*H*HO), 5.12 (d, *J* = 13.0 Hz, 1H, CH*H*O), 4.08 (s, 1H, H-4), 3.63 (m, 1H, H-5), 3.60 (d, *J* = 16.0 Hz, 1H, C*H*HAr), 3.21 (d, *J* = 16.0 Hz, 1H, CH*H*Ar), 2.10 (dd, *J* = 14.0 and 3.0 Hz, 1H, C*H*H-6) and 1.68 (dd, *J* = 14.0 and 3.5 Hz, 1H, CH*H*-6) ppm. ^{13}C NMR (125 MHz, DMSO-d_6) δ 176.9 (C), 150.1 (C), 147.6 (C), 143.1 (C), 140.5 (C), 140.3 (C), 138.4 (C), 137.7 (C), 128.8 (C), 127.3 (CH), 124.7 (2 × CH), 123.9 (CH), 123.4 (CH), 121.8 (CH), 121.4 (CH), 121.2 (CH), 120.5 (C), 120.4 (CH), 74.1 (C), 69.8 (CH), 67.9 (CH), 64.0 (OCH_2), 34.8 (CH_2) and 26.2 (CH_2) ppm. IR (KBr) 3444 (O−H) and 1684 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 573 (MNa^+). HRMS calcd for $C_{25}H_{20}O_6S_2Cl_2Na$ (MNa^+), 572.9971; found, 572.9973.

(1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-2-(benzo-[*b*]thiophen-2-yl)methyl-1,4-dihydroxycyclohex-2-en-1,5-carbolactone (16a). The experimental procedure was the same as for compound **15c** using silyl ether **14a**[28] (75 mg), TBAF (0.29 mL), and THF (1.6 mL). Yield = 50 mg (98%). Light-yellow oil. $[\alpha]_D^{20}$ = −228.5° (*c* 1.0, acetone). ^{1}H NMR (250 MHz, acetone-d_6) δ 7.89 (m, 1H, ArH), 7.76 (m, 2H, 2 × ArH), 7.61 (m, 1H, ArH), 7.39−7.19 (m, 5H, 5 × ArH), 7.12 (m, 1H, ArH), 5.50−5.34 (m, 4H, OCH_2 + 2 × OH), 4.70 (m, 2H, H-4 + H-5), 4.01 (d, *J* = 14.8 Hz, 1H, CH*H*Ar), 3.82 (d, *J* = 14.8 Hz, 1H, C*H*HAr), 2.52 (dd, *J* = 11.0 and 2.8 Hz, 1H, C*H*H-6) and 2.42 (dd, *J* = 11.0 and 5.8 Hz, 1H, CH*H*-6) ppm. ^{13}C NMR (63 MHz, acetone-d_6) δ 177.7 (C), 149.8 (C), 146.4 (C), 143.1 (C), 142.2 (C), 141.9 (C), 141.5 (C), 141.3 (CH), 126.3 (CH), 126.2 (CH), 125.6 (CH), 125.5 (CH), 125.0 (CH), 124.9 (C), 124.6 (CH), 124.5 (CH), 124.2 (CH), 123.7 (CH), 123.3 (CH), 76.9 (CH), 74.3 (C), 67.4 (CH_2), 67.2 (CH), 39.1 (CH_2) and 26.6 (CH_2) ppm. IR (film): 3415 (O−H) and 1788 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 487 (MNa^+). HRMS calcd for $C_{25}H_{20}O_5S_2Na$ (MNa^+), 487.0644; found, 487.0644.

Sodium (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-2-(benzo[*b*]thiophen-2-yl)methyl-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (9a). The experimental procedure was the same as for compound **5c** using lactone **16a** (52 mg), NaOH (220 μL), and THF (1 mL). Yield = 54 mg (97%). Beige solid. $[\alpha]_D^{20} = -62.7°$ (*c* 1.5, MeOH). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.62 (d, *J* = 8.0 Hz, 1H, ArH), 7.87 (m, 1H, ArH), 7.75 (m, 1H, ArH), 7.69 (m, 1H, ArH), 7.55 (m, 1H, ArH), 7.35−7.27 (m, 2H, 2 × ArH), 7.25−7.16 (m, 3H, 3 × ArH), 7.04 (s, 1H, OH), 5.24 (br s, 1H, OH), 5.19−5.12 (m, 3H, OCH_2 + OH), 4.10 (br s, 1H, OH), 3.63 (m, 2H, H-5 + C*H*HAr), 3.23 (d, *J* = 15.2 Hz, 1H, CH*H*Ar), 2.12 (dd, *J* = 14.0 and 3.2 Hz, 1H, C*H*H-6) and 1.70 (dd, *J* = 14.0 and 3.2 Hz, 1H, CH*H*-6) ppm. ^{13}C NMR (63 MHz, DMSO-d_6) δ 177.3 (C), 150.1 (C), 146.3 (C), 142.1 (C), 139.8 (C), 139.2 (C), 139.0 (C), 138.9 (C), 124.2 (CH), 124.1 (CH), 123.6 (CH), 123.4 (CH), 122.7 (CH), 122.4 (CH), 122.2 (CH), 121.9 (CH), 121.8 (CH), 120.8 (CH), 120.5 (C), 74.2 (C), 69.8 (CH), 68.0 (CH), 64.2 (CH_2), 34.9 (CH_2) and 26.2 (CH_2) ppm. IR (KBr): 3398 (O−H) and 1601 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 505 (MH^+). HRMS calcd for $C_{25}H_{22}O_6S_2Na$ (MH^+), 505.0750; found, 505.0751.

(1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(5-methylbenzo[*b*]thiophen-2-yl)methoxy-2-(5-methylbenzo[*b*]thiophen-2-yl)methylcyclohex-2-en-1,5-carbolactone (16b). The experimental procedure was the same as for compound **15c** using silyl ether **14b**[28] (119 mg), TBAF (0.44 mL), and THF (2.4 mL). Yield = 70 mg (84%). Beige solid. $[\alpha]_D^{20} = -232.4°$ (*c*1.7, acetone). ^{1}H NMR (250 MHz, acetone-d_6) δ 7.74 (d, *J* = 8.3 Hz, 1H, ArH), 7.62 (d, *J* = 8.3 Hz, 1H, ArH), 7.52 (s, 1H, ArH), 7.35 (s, 1H, ArH), 7.24 (s, 1H, ArH), 7.17 (dd, *J* = 8.0 and 0.8 Hz, 1H, ArH), 7.06 (dd, *J* = 8.0 and 0.8 Hz, 1H, ArH), 6.99 (s, 1H, ArH), 5.43 (d, *J* = 12.5 Hz, 1H, OC*H*H), 5.35 (d, *J* = 12.5 Hz, 1H, OCH*H*), 4.68 (m, 2H, H-4+H-5), 3.97 (d, *J* = 14.8 Hz, 1H, C*H*HAr), 3.79 (d, *J* = 14.8 Hz, 1H, CH*H*Ar), 2.51 (d, *J* = 12.5 Hz, 1H, CH*H*-6), 2.45−2.38 (m, 1H, C*H*H-6), 2.41 (s, 3H, Me) and 2.38 (s, 3H, Me) ppm. ^{13}C NMR (63 MHz, acetone-d_6) δ 177.8 (C), 149.8 (C), 146.5 (C), 143.1 (C), 142.5 (C), 141.7 (C), 139.2 (C), 138.7 (C), 135.7 (C), 135.1 (C), 127.9 (CH), 126.6 (CH), 125.4 (CH), 125.0 (CH), 124.5 (CH), 124.4 (CH), 123.8 (CH), 123.3 (CH), 123.1 (CH), 76.9 (CH), 74.3 (C), 67.4 (OCH_2), 67.2 (CH), 39.1 (CH_2), 26.6 (CH_2) and 22.3 (2 × CH_3) ppm. IR (film): 3471 (O−H), 3344 (O−H) and 1770 (C═O) cm^{-1}. MS (CI) *m*/*z* (%) 493 (MH^+). HRMS calcd for $C_{27}H_{25}O_5S_2$ (MH^+), 493.1143; found, 493.1131.

Sodium (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(5-methylbenzo[*b*]thiophen-2-yl)methoxy-2-(5-methylbenzo[*b*]thiophen-2-yl)methylcyclohex-2-en-1-carboxylate (9b). The experimental procedure was the same as for compound **5c** using carbolactone **16b** (35 mg), NaOH (126 μL), and THF (0.65 mL). Yield = 37 mg (98%). Beige solid. $[\alpha]_D^{20} = -61.3°$ (*c* 1.5, MeOH). ^{1}H NMR (250 MHz, DMSO-d_6) δ 8.63 (d, *J* = 8.0 Hz, 1H, ArH), 7.73 (d, *J* = 8.0 Hz, 1H, ArH), 7.61 (d, *J* = 8.0 Hz, 1H, ArH), 7.45 (br s, 1H, ArH), 7.29 (br s, 1H, ArH), 7.13 (dd, *J* = 8.0 and 1.6 Hz, 1H, ArH), 7.08 (br s, 1H, ArH), 7.01 (dd, *J* = 8.0 and 1.6 Hz, 1H, ArH), 6.92 (s, 1H, OH), 5.21 (br s, 1H, OH), 5.15 (br s, 1H, OH), 5.14 (d, *J* = 13.2 Hz, 1H, OC*H*H), 5.10 (d, *J* = 13.2 Hz, 1H, OCH*H*), 4.08 (br s, 1H, H-4), 3.64 (m, 1H, H-5), 3.60 (d, *J* = 15.2 Hz, 1H, C*H*HAr), 3.19 (d, *J* = 15.2 Hz, 1H, CH*H*Ar), 2.38 (s, 3H, Me), 2.35 (s, 3H, Me), 2.11 (dd, *J* = 14.0 and 3.2 Hz, 1H, CH*H*-6) and 1.68 (dd, *J* = 14.0 and 3.2 Hz, 1H, C*H*H-6) ppm. ^{13}C NMR (100 MHz, DMSO-d_6) δ 177.2 (C), 150.1 (C), 146.4 (C), 142.1 (C), 140.1 (C), 139.4 (C), 136.4 (C), 136.1 (C), 133.3 (C), 132.5 (C), 125.7 (CH), 124.3 (CH), 123.3 (CH), 122.2 (CH), 122.0 (CH), 121.7 (CH), 121.4 (CH), 120.6 (CH), 120.5 (C), 74.3 (C), 69.8 (CH), 68.0 (CH), 64.2 (OCH_2), 34.9 (CH_2), 26.3 (CH_2), 21.0 (CH_3) and 20.9 (CH_3) ppm. IR (KBr): 3435 (O−H) and 1599 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 533 (MH^+). HRMS calcd for $C_{27}H_{26}O_6S_2Na$ (MH^+), 533.1068; found, 533.1072. Elemental analysis $C_{27}H_{25}O_6S_2Na \cdot 2H_2O$ Calcd: C, 57.03; H, 5.14; S, 11.28. Found: C, 56.69; H, 5.19; S, 10.91.

(1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-yl)methoxy-2-(benzo[*b*]thiophen-5-yl)methyl-1,4-dihydroxycyclohex-2-en-1,5-carbolactone (16d). The experimental procedure was the same as for compound **5c** using ether **14d**[28] (97 mg, 0.14 mmol), TBAF (280 μL), and THF (2.0 mL). Yield = 56 mg (86%). White solid; mp 123−125 °C. $[\alpha]_D^{20} = -163°$ (*c* 1.5, acetone). ^{1}H NMR (250 MHz, CD_3OD) δ 7.74 (d, *J* = 8.2 Hz, 1H, ArH), 7.61 (m, 3H, 3 × ArH), 7.49 (d, *J* = 5.5 Hz, 1H, ArH), 7.39 (d, *J* = 5.5 Hz, 1H, ArH), 7.19 (m, 3H, 3 × ArH), 7.10 (d, *J* = 5.5 Hz, 1H, ArH), 5.13 (d, *J* = 11.5 Hz, 1H, OC*H*H), 4.92 (d, *J* = 11.5 Hz, 1H, OCH*H*), 4.57 (m, 1H, H-5), 4.51 (d, *J* = 3.2 Hz, 1H, H-4), 3.73 (d, *J* = 14.2 Hz, 1H, C*H*HAr), 3.56 (d, *J* = 14.2 Hz, 1H, CH*H*Ar) and 2.30 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, acetone-d_6) δ 177.8 (C), 149.6 (C), 141.4 (C), 140.6 (C), 138.7 (C), 138.4 (C), 135.8 (C), 128.7 (CH), 127.7 (CH), 127.5 (CH), 125.8 (CH), 125.4 (CH), 125.3 (C), 125.3 (CH), 125.2 (CH), 125.1 (C), 124.2 (CH), 123.8 (CH), 123.1 (CH), 76.6 (CH), 74.2 (C), 71.4 (CH_2), 66.8 (CH), 38.9 (CH_2) and 30.9 (CH_2) ppm. IR (KBr) 3452 (O−H), 3363 (O−H) and 1770 (C═O) cm^{-1}. MS (CI) *m*/*z* (%) 465 (MH^+). HRMS calcd for $C_{25}H_{21}O_5S_2$ (MH^+), 465.0830; found, 465.0831.

Sodium (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-yl)methoxy-2-(benzo[*b*]thiophen-5-yl)methyl-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (9d). The experimental procedure was the same as for compound **5c** using lactone **16d** (25 mg), NaOH (0.1 mL), and THF (0.4 mL). Yield = 25 mg (99%). White solid; mp 197−200 °C (dec). $[\alpha]_D^{20} = -67°$ (*c* 1.3, 50% MeOH/H_2O). ^{1}H NMR (250 MHz, 50% CD_3OD/D_2O) δ 7.66 (m, 3H, 3 × ArH), 7.47 (m, 2H, 2 × ArH), 7.26 (d, *J* = 8.2 Hz, 1H, ArH), 7.14 (m, 2H, 2 × ArH), 7.05 (d, *J* = 5.5 Hz, 1H, ArH), 6.98 (d, *J* = 8.2 Hz, 1H, ArH), 4.84 (d, *J* = 10.5 Hz, 1H, OC*H*H), 4.58 (d, *J* = 10.5 Hz, 1H, OCH*H*), 4.41 (d, *J* = 5.0 Hz, 1H, H-4), 3.98 (m, 1H, H-5), 3.57 (d, *J* = 15.7 Hz, 1H, C*H*HAr), 3.28 (d, *J* = 15.7 Hz, 1H, CH*H*Ar) and 2.20 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, 50% CD_3OD/D_2O) δ 181.9 (C), 153.1 (C), 141.9 (C), 141.6 (C), 141.0 (C), 139.7 (C), 138.8 (C), 135.3 (C), 128.9 (CH), 128.2 (CH), 127.6 (CH), 126.4 (CH), 125.8 (CH), 125.7 (C), 125.3 (CH), 125.0 (CH), 124.3 (CH), 123.9 (CH), 123.5 (CH), 78.5 (C), 72.2 (CH), 72.0 (CH_2), 70.9 (CH), 38.8 (CH_2) and 33.8 (CH_2) ppm. IR (KBr) 3410 (O−H) and 1595 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 481 (M^-). HRMS calcd for $C_{25}H_{21}O_6S_2Na$ (M^-), 481.0774; found, 481.0776.

(1*R*,4*S*,5*R*)-1,4-Di(*tert*-butyldimethylsilyloxy)-3-(naphth-2-yl)methoxycyclohex-2-en-1,5-carbolactone (13e) and (1*R*,4*S*,5*R*)-1,4-Di(*tert*-butyldimethylsilyloxy)-3-(naphth-2-yl)methoxy-2-(naphth-2-yl)methylcyclohex-2-en-1,5-carbolactone (14e). The experimental procedure was the same as for compounds **13c** and **14c** using ketone **12**[22] (522 mg, 1.30 mmol), KHMDS (5.2 mL), and 2-(bromomethyl)naphtalene (577 mg). Purification by flash chromatography on silica gel eluting with diethyl ether/hexanes [(1) 5:95; (2) 10:90] gave ethers **13e** (302 mg, 43%) and **14e** (259 mg, 29%), both as colorless oils.

Data for **13e**: $[\alpha]_D^{20} = -106°$ (*c* 1.0, $CHCl_3$). ^{1}H NMR (250 MHz, $CDCl_3$) δ 7.85 (m, 4H, 4 × ArH), 7.49 (m, 3H, 3 × ArH), 5.11 (s, 1H, H-2), 4.93 (d, 1H, *J* = 11.5 Hz, C*H*HO), 4.86 (d, 1H, *J* = 11.5 Hz, CH*H*O), 4.54 (dd, 1H, *J* = 5.0 and 3.5 Hz, H-5), 4.27 (d, 1H, *J* = 3.5 Hz, H-4), 2.47 (d, 1H, *J* = 10.7 Hz, H-6_{eq}), 2.38 (dd, 1H, *J* = 10.7 and 5.0 Hz, H-6_{ax}), 0.99 (s, 9H, $C(CH_3)_3$), 0.95 (s, 9H, $C(CH_3)_3$), 0.21 (s, 3H, $SiCH_3$), 0.16 (s, 3H, $SiCH_3$), 0.14 (s, 3H, $SiCH_3$) and 0.12 (s, 3H, $SiCH_3$) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ 176.0 (C), 153.4 (C), 133.2 (C), 133.0 (C), 132.9 (C), 128.1 (CH), 127.7 (CH), 127.6 (CH), 126.5 (CH), 126.1 (CH), 126.0 (CH), 125.3 (CH), 104.9 (CH), 75.2 (CH), 73.7 (C), 69.6 (OCH_2), 67.4 (CH), 38.0 (CH_2), 25.6 ($C(CH_3)_3$), 25.5 ($C(CH_3)_3$), 18.0 ($C(CH_3)_3$), 17.9 ($C(CH_3)_3$), −3.2 (2 × CH_3), −4.6 (CH_3) and −5.2 (CH_3) ppm. IR (film) 1803 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 563 (MNa^+). HRMS calcd for $C_{30}H_{44}O_5Si_2Na$ (MNa^+), 563.2619; found, 563.2622.

Data for **14e**: $[\alpha]_D^{20} = -151°$ (*c* 1.0, $CHCl_3$). ^1H NMR (250 MHz, $CDCl_3$) δ 7.84−7.60 (m, 7H, 7 × ArH), 7.51−7.26 (m, 7H, 7 × ArH), 4.98 (d, 1H, *J* = 11.5 Hz, C*H*HO), 4.87 (d, 1H, *J* = 11.5 Hz, CH*H*O), 4.65 (dd, 1H, *J* = 5.7 and 3.5 Hz, H-5), 4.56 (d, 1H, *J* = 3.5 Hz, H-4), 3.94 (d, 1H, *J* = 15.0 Hz, C*H*HAr), 3.80 (d, 1H, *J* = 15.0 Hz, CH*H*Ar), 2.67 (d, 1H, *J* = 10.7 Hz, H-6_{eq}), 2.52 (dd, 1H, *J* = 10.7 and 6.0 Hz, H-6_{ax}), 1.00 (s, 9H, $C(CH_3)_3$), 0.76 (s, 9H, $C(CH_3)_3$), 0.22 (s, 3H, $SiCH_3$), 0.21 (s, 3H, $SiCH_3$), 0.19 (s, 3H, $SiCH_3$) and 0.08 (s, 9H, $SiCH_3$) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ 175.6 (C), 149.0 (C), 137.6 (C), 134.3 (C), 133.4 (C), 133.1 (C), 132.8 (C), 131.9 (C), 129.1 (C), 128.0 (CH), 127.9 (CH), 127.6 (CH), 127.5 (CH), 127.4 (CH), 127.4 (CH), 127.3 (CH), 126.6 (CH), 126.0 (CH), 126.0 (CH), 125.9 (CH), 125.5 (CH), 125.0 (CH), 124.8 (CH), 75.0 (C), 74.8 (CH), 72.7 (OCH_2), 67.3 (CH), 37.6 (CH_2), 30.4 (CH_2), 25.7 ($C(CH_3)_3$), 25.4 ($C(CH_3)_3$), 18.0 (2 × $C(CH_3)_3$), −3.3 (CH_3), −3.5 (CH_3), −4.5 (CH_3) and −4.5 (CH_3) ppm. IR (Film) 1799 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 703 (MNa^+). HRMS calcd for $C_{41}H_{52}O_5Si_2Na$ (MNa^+), 703.3245; found, 703.3248.

(1*R*,4*S*,5*R*)-1,4-Dihydroxy-3-(naphth-2-yl)methoxycyclohex-2-en-1,5-carbolactone (15e). The experimental procedure was the same as for compound **15c** using lactone **13e** (220 mg), TBAF (0.9 mL), and THF (5.8 mL). Yield = 88 mg (69%). Colorless oil. $[\alpha]_D^{20} = -112°$ (*c* 1.0, MeOH). ^1H NMR (250 MHz, CD_3OD) δ 7.79 (m, 4H, 4 × ArH), 7.44 (m, 3H, 3 × ArH), 5.01 (s, 1H, H-2), 4.91 (s, 2H, OCH_2Ar), 4.59 (m, 1H, H-5), 4.11 (d, *J* = 3.5 Hz, 1H, H-4) and 2.27 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 179.3 (C), 155.8 (C), 135.3 (C), 134.8 (C), 134.6 (C), 129.2 (CH), 129.0 (CH), 128.7 (CH), 127.6 (CH), 127.3 (CH), 127.2 (CH), 126.6 (CH), 105.1 (CH), 77.0 (CH), 73.0 (C), 70.9 (CH_2), 67.8 (CH) and 38.4 (CH_2) ppm. IR (film) 3446 (O−H) and 1770 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 335 (MNa^+). HRMS calcd for $C_{18}H_{16}O_5Na$ (MNa^+), 335.0890; found, 335.0889.

(1*R*,4*S*,5*R*)-1,4-Dihydroxy-3-(naphth-2-yl)methoxy-2-(naphth-2-yl)methylcyclohex-2-en-1,5-carbolactone (16e). The experimental procedure was the same as for compound **15c** using lactone **14e** (73 mg), TBAF (0.24 mL), and THF (1.6 mL). Yield = 32 mg (64%). White solid; mp 203−207 °C. $[\alpha]_D^{20} = -190°$ (*c* 1.4, acetone). ^1H NMR (250 MHz, acetone-d_6) δ 7.89−7.65 (m, 8H, 8 × ArH), 7.53−7.37 (m, 6H, 6 × ArH), 5.33 (d, *J* = 11.8 Hz, 1H, OC*H*H), 5.15 (d, *J* = 11.8 Hz, 1H, OCH*H*),4.69 (m, 2H, H-4 + H-5), 3.92 (d, *J* = 14.5 Hz, 1H, C*H*HAr), 3.74 (d, *J* = 14.5 Hz, 1H, CH*H*Ar), 2.53 (d, *J* = 11.0 Hz, 1H, H-6_{eq}) and 2.41 (dd, 1*J* = 11.0 and 5.7 Hz, 1H, H-6_{ax}) ppm. ^{13}C NMR (63 MHz, acetone-d_6) δ 177.8 (C), 149.8 (C), 140.1 (C), 140.1 (C), 137.1 (C), 135.2 (C), 134.9 (C), 134.6 (C), 133.6 (C), 129.5 (CH), 129.5 (CH), 129.4 (CH), 129.1 (CH), 129.0 (CH), 128.9 (CH), 128.7 (CH), 128.4 (CH), 127.7 (CH), 127.6 (CH), 127.5 (CH), 127.2 (CH), 127.0 (CH), 126.4 (CH), 76.7 (CH), 74.3 (C), 71.4 (CH_2), 66.9 (CH), 38.9 (CH_2) and 31.2 (CH_2) ppm. IR (KBr) 3460 (O−H), 3346 (O−H) and 1768 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 475 (MNa^+). HRMS calcd for $C_{29}H_{24}O_5Na$ (MNa^+), 475.1516; found, 475.1511.

Sodium (1*R*,4*S*,5*R*)-1,4-Trihydroxy-3-(naphth-2-yl)methoxycyclohex-2-en-1-carboxylate (5e). The experimental procedure was the same as for compound **5c** using carbolactone **15e** (8 mg, 0.02 mmol), NaOH (51 μL), and THF (0.2 mL). Yield = 9 mg (99%). Beige solid; mp 54 °C (dec). $[\alpha]_D^{20} = -12°$ (*c* 1.1, H_2O). ^1H NMR (300 MHz, 50% CD_3OD/D_2O) δ 7.93 (m, 4H, 4 × ArH), 7.57 (m, 3H, 3 × ArH), 5.04 (d, *J* = 11.7 Hz, 1H, OC*H*H), 4.97 (d, *J* = 11.7 Hz, 1H, OCH*H*), 4.95 (s, 1H, H-2), 4.09 (d, *J* = 6.0 Hz, 1H, H-4), 4.01 (m, 1H, H-5), 2.16 (dd, *J* = 13.8 and 8.1 Hz, 1H, H-6_{ax}) and 2.06 (dd, *J* = 13.8 and 3.9 Hz, 1H, H-6_{eq}) ppm. ^{13}C NMR (75 MHz, 50% CD_3OD/D_2O) δ 182.1 (C), 157.0 (C), 135.4 (C), 134.3 (C), 134.1 (C), 129.2 (CH), 128.9 (CH), 128.6 (CH), 127.5 (CH), 127.4 (CH), 127.3 (CH), 126.8 (CH), 102.2 (CH), 74.8 (CH), 72.8 (C), 70.9 (CH), 70.6 (CH_2) and 38.7 (CH_2) ppm. IR (KBr) 3435 (O−H) and 1660 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 375 (MNa^+). HRMS calcd for $C_{18}H_{17}O_6Na_2$ (MNa^+), 375.0815; found, 375.0817.

Sodium (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(naphth-2-yl)-methoxy-2-(naphth-2-yl)methylcyclohex-2-en-1-carboxylate (9e). The experimental procedure was the same as for compound **5c** using carbolactone **16e** (29 mg), NaOH (120 μL), and THF (0.6 mL). Yield = 28 mg (95%). White solid; mp 184−187 °C $[\alpha]_D^{20} = -4°$ (*c*1.1, H_2O). ^1H NMR (250 MHz, 50% D_2O/CD_3CN) δ 7.76−7.57 (m, 6H, 6 × ArH), 7.46−7.29 (m, 7H, 7 × ArH), 7.15 (br d, *J* = 8.2 Hz, 1H, ArH), 4.92 (d, *J* = 11.2 Hz, 1H, OC*H*H), 4.71 (d, *J* = 11.2 Hz, 1H, OCH*H*), 4.33 (d, *J* = 3.0 Hz, 1H, H-4), 3.88 (m, 1H, H-5), 3.60 (d, *J* = 15.7 Hz, 1H, C*H*HAr), 3.22 (d, *J* = 15.7 Hz, 1H, CH*H*Ar) and 2.10 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, 50% D_2O/CD_3CN) δ 180.5 (C), 151.9 (C), 140.1 (C), 136.0 (C), 134.2 (C), 133.7 (C), 133.5 (C), 132.5 (C), 128.7 (CH), 128.6 (CH), 128.5 (CH), 128.3 (CH), 128.2 (2 × CH), 127.9 (CH), 127.3 (CH), 127.1 (CH), 126.9 (CH), 126.8 (CH), 126.8 (CH), 126.5 (CH), 125.7 (CH), 122.3 (C), 76.9 (C), 70.8 (CH), 70.4 (CH_2), 69.2 (CH), 36.8 (CH_2) and 32.7 (CH_2) ppm. IR (KBr) 3435 (O−H) and 1660 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 493 (MH^+). HRMS calcd for $C_{29}H_{26}O_6Na$ (MH^+), 493.1622; found, 493.1620.

(1*R*,4*S*,5*R*)-1,4-Dihydroxy-3-(thien-3-yl)methoxy-2-(thien-3-yl)methylcyclohex-2-en-1,5-carbolactone (16f). The experimental procedure was the same as for compound **15c** using silyl ether **14f**[28] (145 mg), TBAF (0.54 mL), and THF (3.5 mL). Yield = 75 mg (86%). White solid; mp 95−97 °C. $[\alpha]_D^{20} = -189°$ (*c* 1.2, MeOH). ^1H NMR (300 MHz, CD_3OD) δ 7.36 (d, *J* = 4.8 Hz, 1H, ArH), 7.07 (m, 1H, ArH), 7.02 (s, 1H, ArH), 6.96 (m, 1H, ArH), 6.81 (s, 2H, 2 × ArH), 5.22 (d, *J* = 12.3 Hz, 1H, OC*H*H), 5.11 (d, *J* = 12.3 Hz, 1H, OCH*H*), 4.61 (m, 1H, H-5), 4.51 (d, *J* = 2.4 Hz, 1H, H-4), 3.76 (d, *J* = 14.4 Hz, 1H, C*H*HAr), 3.64 (d, *J* = 14.4 Hz, 1H, CH*H*Ar), 2.38 (d, *J* = 11.1 Hz, 1H, H-6_{ax}) and 2.32 (dd, *J* = 11.1 and 6.0 Hz, 1H, H-6_{eq}) ppm. ^{13}C NMR (75 MHz, CD_3OD) δ 178.8 (C), 148.5 (C), 144.1 (C), 141.0 (C), 127.9 (CH), 127.7 (CH), 127.3 (CH), 127.3 (CH), 126.2 (CH + C), 124.0 (CH), 76.8 (CH), 73.6 (C), 66.1 (CH), 65.9 (CH_2), 38.4 (CH_2) and 24.9 (CH_2) ppm. IR (KBr) 3498 (O−H), 3413 (O−H) and 1774 (C═O) cm^{-1}. MS (CI) *m*/*z* (%) 365 (MH^+). HRMS calcd for $C_{17}H_{16}O_5S_2$ (MH^+), 365.0517; found, 365.0517.

Sodium (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(thien-3-yl)methoxy-2-(thien-3-yl)methylcyclohex-2-en-1-carboxylate (9f). The experimental procedure was the same as for compound **5c** using lactone **16f** (25 mg), NaOH (140 μL), and THF (0.6 mL). Yield = 25 mg (97%). Beige solid; mp 178−181 °C. $[\alpha]_D^{20} = -89°$ (*c* 1.2, H_2O). ^1H NMR (300 MHz, D_2O) δ 7.46 (d, *J* = 4.8 Hz, 1H, ArH), 7.19 (dt, *J* = 5.1 and 1.2 Hz, 1H, ArH), 7.13 (br d, *J* = 2.7 Hz, 1H, ArH), 7.05 (ddd, *J* = 5.1, 3.6, and 0.6 Hz, 1H, ArH), 6.92 (dt, *J* = 3.6 and 0.9 Hz, 1H, ArH), 6.85 (m, 1H, ArH), 5.18 (d, *J* = 12.0 Hz, 1H, C*H*HO), 4.92 (d, *J* = 12.0 Hz, 1H, CH*H*O), 4.37 (d, *J* = 6.6 Hz, 1H, H-4), 3.93 (m, 1H, H-5), 3.68 (d, *J* = 15.6 Hz, 1H, C*H*HAr), 3.28 (d, *J* = 15.6 Hz, 1H, CH*H*Ar), 2.16 (dd, *J* = 13.8 and 11.4 Hz, 1H, H-6_{ax}) and 2.03 (dd, *J* = 13.8 and 3.3 Hz, 1H, H-6_{eq}) ppm. ^{13}C NMR (75 MHz, D_2O) δ 182.9 (C), 153.8 (C), 146.8 (C), 142.0 (C), 131.2 (CH), 130.3 (CH), 130.2 (CH), 129.8 (CH), 128.4 (CH), 126.8 (CH), 126.7 (C), 79.8 (C), 73.0 (CH), 72.9 (CH), 68.4 (CH_2), 41.7 (CH_2) and 29.8 (CH_2) ppm. IR (KBr) 3390 (O−H) and 1597 (C═O) cm^{-1}. EM (ESI) *m*/*z* (%) 381 (M^-). HRMS calcd for $C_{17}H_{17}O_6S_2$ (M^-), 381.0461; found, 381.0461.

(1*R*,4*S*,5*R*)-2-Allyl-1,4-di(*tert*-butyldimethylsilyloxy)-3-(benzo[*b*]thiophen-2-yl)methoxycyclohex-2-en-1,5-carbolactone (18g). To a solution of KHMDS (1.8 mL, 0.91 mmol, 0.5 M in toluene) in dry DMF (3 mL), under argon and at −78 °C, was added a solution of (2*S*)-2-allyl ketone **17**[32] (200 mg, 0.45 mmol) in dry DMF (3 mL) and dry toluene (1.9 mL). The resultant solution was stirred at this temperature for 30 min. A solution of 2-(bromomethyl)benzo[*b*]thiophene[27] (206 mg, 0.91 mmol) in DMF (1.8 mL) and toluene (1.2 mL), both dry, was then added. After 1 h, water and brine were added. The aqueous phase was extracted with diethyl ether (3 × 2 mL). The combined organic extracts were dried (anhyd $MgSO_4$), filtered, and concentrated. The resulting residue was purified

by flash chromatography on silica gel, eluting with diethyl ether/hexanes [1) (0:100), 2) (20:80)] to afford ether **18g** (118 mg, 45%) as a pale-yellow oil. $[\alpha]_D^{20} = -100°$ (*c* 1.0, $CHCl_3$). ^1H NMR (250 MHz, $CDCl_3$) δ 7.66 (dd, *J* = 6.5 and 2.2 Hz, 1H, ArH), 7.59 (dd, *J* = 6.5 and 2.5 Hz, 1H, ArH), 7.17 (m, 2H, 2 × ArH), 7.03 (s, 1H, ArH), 5.87−5.71 (m, 1H, C*H*=CH_2), 4.95−4.80 (m, 4H, OCH_2 + CH=CH_2), 4.34 (dd, *J* = 4.7 and 3.5 Hz, 1H, H-5), 4.17 (d, *J* = 3.5 Hz, 1H, H-4), 2.96 (d, *J* = 6.2 Hz, 2H, CH_2-CH=CH_2), 2.26 (m, 2H, CH_2-6), 0.78 (s, 9H, $C(CH_3)_3$), 0.77 (s, 9H, $C(CH_3)_3$), 0.08 (s, 3H, $SiCH_3$), 0.02 (s, 3H, $SiCH_3$), 0.00 (s, 3H, $SiCH_3$) and −0.03 (s, 3H, $SiCH_3$) ppm. ^{13}C NMR (75 MHz, $CDCl_3$) δ 176.3 (C), 148.3 (C), 140.9 (C), 139.2 (C), 136.5 (CH), 131.2 (C), 125.1 (CH), 125.0 (CH), 124.3 (CH), 123.1 (CH), 123.0 (CH), 122.9 (C), 116.1 (CH_2), 77.3 (C), 75.4 (CH), 69.8 (CH_2), 68.1 (CH), 38.0 (CH_2), 30.1 (CH_2), 26.4 ($C(CH_3)_3$), 26.2 ($C(CH_3)_3$), 18.9 ($C(CH_3)_3$), 18.7 ($C(CH_3)_3$), −2.5 ($SiCH_3$), −2.6 ($SiCH_3$), −3.8 ($SiCH_3$) and −4.0 ($SiCH_3$) ppm. IR (film): 1799 (C=O) cm^{-1}. MS (CI) *m*/*z* (%) 587 (MH^+). HRMS calcd for $C_{31}H_{47}O_5SSi_2$ (MH^+), 587.2683; found, 587.2682.

(1*R*,4*S*,5*R*)-2-Allyl-1,4-dihydroxy-3-(benzo[*b*]thiophen-2-yl)methoxycyclohex-2-en-1,5-carbolactone (19g). The experimental procedure was the same as for compound **15c** using silyl ether **18g** (78 mg), TBAF (0.29 mL), and THF (0.9 mL). Yield = 38 mg (83%). Beige solid; mp 122−125 °C. $[\alpha]_D^{20} = -143°$ (*c* 1.5, MeOH). ^1H NMR (250 MHz, CD_3OD) δ 7.76 (m, 1H, ArH), 7.69 (m, 1H, ArH), 7.26 (m, 2H, 2 × ArH), 7.22 (s, 1H, ArH), 5.78 (m, 1H, C*H*=CH_2), 5.22 (d, *J* = 12.5 Hz, 1H, OC*H*H), 5.12 (d, *J* = 12.5 Hz, 1H, OCH*H*), 4.97 (dq, *J* = 17.0 and 1.7 Hz, 1H, CH=C*H*H), 4.81 (m, 1H, CH=CH*H*), 4.55 (m, 1H, H-5), 4.40 (d, *J* = 3.5 Hz, 1H, H-4), 3.00 (d, *J* = 6.5 Hz, 2H, CH_2CH=CH_2) and 2.29 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 178.8 (C), 148.1 (C), 142.3 (C), 141.5 (C), 140.8 (C), 137.1 (CH), 126.5 (C), 125.5 (CH), 125.3 (CH), 124.7 (CH), 123.8 (CH), 123.2 (CH), 115.5 (CH_2), 76.8 (CH), 73.8 (C), 67.2 (CH_2), 66.4 (CH_2), 38.3 (CH_2) and 29.5 (CH_2) ppm. IR (KBr): 3482 (O−H), 3369 (O−H) and 1780 (C=O) cm^{-1}. MS (ESI) *m*/*z* (%) 381 (MNa^+). HRMS calcd for $C_{19}H_{18}O_5SNa$ (MNa^+), 381.0751; found, 381.0758.

Sodium (1*R*,4*S*,5*R*)-2-Allyl-3-(benzo[*b*]thiophen-2-yl)-methoxy-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (9g). The experimental procedure used was the same as for compound **5c** using carbolactone **19g** (30 mg), NaOH (160 μL), and THF (0.75 mL). Yield = 33 mg (99%). Beige solid. $[\alpha]_D^{20} = -49°$ (*c* 1.1, H_2O). ^1H NMR (250 MHz, D_2O) δ 7.80 (m, 2H, 2 × ArH), 7.34 (m, 2H, 2 × ArH), 7.30 (s, 1H, ArH), 5.79 (m, 1H, C*H*=CH_2), 5.09 (d, *J* = 11.8 Hz, 1H, OC*H*H), 4.98 (m, 3H, OCH*H* + CH=CH_2), 4.32 (d, *J* = 6.7 Hz, 1H, H-4), 3.93 (m, 1H, H-5), 2.91 (dd, *J* = 15.0 and 6.5 Hz, 1H, CH*H*CH=CH_2), 2.66 (dd, *J* = 15.0 and 6.5 Hz, 1H, C*H*HCH=CH_2) and 2.05 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, D_2O) δ 180.5 (C), 150.9 (C), 140.8 (C), 140.7 (C), 139.8 (C), 137.4 (CH), 125.4 (CH), 125.2 (CH), 124.6 (CH), 123.9 (C), 123.1 (CH), 115.9 (CH_2), 77.1 (C), 70.7 (CH), 70.6 (CH), 39.4 (CH_2) and 31.5 (CH_2) ppm. IR (KBr): 3427 (O−H) and 1597 (C=O) cm^{-1}. MS (ESI) *m*/*z* (%) 399 (MH^+). HRMS calcd for $C_{19}H_{20}O_6SNa$ (MH^+), 399.0873; found, 399.0887.

(1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-1,4-di(*tert*-butyldimethylsilyloxy)-2-(thien-2-yl)methylcyclohex-2-en-1,5-carbolactone (18h). A solution of ketone **12**[22] (500 mg, 1.25 mmol) in dry THF (12.5 mL), at room temperature and under argon, was treated with a solution of LHMDS (1.9 mL, 1.87 mmol, 1 M in THF). The resulting mixture was stirred for 1 h and was then treated with a solution of 2-iodomethylthiophene[28] (560 mg, 2.5 mmol) in dry THF (4 mL). After 30 min, the solvent was removed and the residue was purified by flash chromatography on silica gel, eluting with diethyl ether/hexanes (5:95) to yield a diastereomeric mixture of *C*-alkylated products (206 mg, 33%). A solution of 50 mg (0.10 mmol) of the latter mixture in DMF (0.5 mL) and toluene (0.5 mL), both dry, was treated with a solution of KHMDS (0.4 mL, 0.5 M in toluene) in dry DMF (0.2 mL). After 20 min, there was a solution of 2-(bromomethyl)-benzo[*b*]thiophene[28] (46 mg, 0.20 mmol) in dry DMF (0.5 mL) and dry toluene (0.3 mL). After 40 min, water and diethyl ether were added, the organic layer was separated, and the aqueous layer was extracted with diethyl ether (×3). The combined organic extracts were dried (anhyd Na_2SO_4), filtered, and concentrated under reduced pressure. The resulting residue was purified by flash chromatography, eluting with a gradient of diethyl ether/hexanes [(1) 0:100, (2) 5:95] to afford compound **18h** (28 mg, 44%) as a yellow oil. $[\alpha]_D^{20} = -132°$ (*c* 1.2, $CHCl_3$). ^1H NMR (250 MHz, CD_3OD) δ 7.80 (m, 1H, ArH), 7.72 (m, 1H, ArH), 7.32 (m, 2H, 2 × ArH), 7.07 (m, 2H, 2 × ArH), 6.86 (dd, *J* = 3.5 and 5.0 Hz, 1H, ArH), 6.79 (d, *J* = 3.5 Hz, 1H, ArH), 5.03 (s, 2H, OCH_2), 4.54 (dd, *J* = 5.8 and 3.5 Hz, 1H, H-5), 4.44 (d, *J* = 3.5 Hz, 1H, H-4), 3.87 (d, *J* = 15.2 Hz, 1H, C*H*HAr), 3.74 (d, *J* = 15.2 Hz, 1H, CH*H*Ar), 2.53 (d, *J* = 10.8 Hz, 1H, H-6_{ax}), 2.42 (dd, *J* = 10.8 and 5.8 Hz, 1H, H-6_{eq}), 0.95 (s, 9H, $C(CH_3)_3$), 0.81 (s, 9H, $C(CH_3)_3$), 0.20 (s, 3H, CH_3), 0.18 (s, 3H, CH_3), 0.16 (s, 3H, CH_3) and 0.06 (s, 3H, CH_3) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ 175.2 (C), 148.2 (C), 142.7 (C), 140.0 (C), 139.9 (C), 139.1 (C), 129.7 (C), 126.4 (CH), 124.6 (CH), 124.3 (CH), 124.2 (CH), 123.6 (CH), 122.8 (CH), 122.6 (CH), 122.3 (CH), 74.6 (C), 74.5 (CH), 68.5 (CH_2), 67.3 (CH), 37.4 (CH_2), 25.6 ($C(CH_3)_3$), 24.7 ($C(CH_3)_3$), 18.0 ($C(CH_3)_3$), 17.9 ($C(CH_3)_3$), −3.4 (CH_3), −3.5 (CH_3), −4.5 (CH_3) and −4.6 (CH_3) ppm. IR (film) 1797 (C=O) cm^{-1}. MS (ESI) *m*/*z* (%) 665 (MNa^+). HRMS calcd for $C_{33}H_{46}O_5S_2Si_2$ Na(MNa^+), 665.2217; found, 665.2225.

(1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-1,4-dihydroxy-2-(thien-2-yl)methylcyclohex-2-en-1,5-carbolactone (19h). The experimental procedure was the same as for compound **15c** using silyl ether **18h** (15 mg), TBAF (50 μL), and THF (0.4 mL). Yield = 7 mg (84%). White solid. $[\alpha]_D^{20} = -155°$ (*c* 1.2, MeOH). ^1H NMR (250 MHz, CD_3OD) δ 7.71 (m, 2H, 2 × ArH), 7.25 (m, 3H, 3 × ArH), 7.18 (s, 1H, ArH), 7.00 (dd, *J* = 5.0 and 1.5 Hz, 1H, ArH), 6.74 (m, 1H, ArH), 5.27 (d, *J* = 12.5 Hz, 1H, OC*H*H), 5.17 (d, *J* = 12.5 Hz, 1H, OCH*H*), 4.55 (m, 1H, H-5), 4.47 (d, *J* = 3.2 Hz, 1H, H-4), 3.78 (d, *J* = 14.8 Hz, 1H, C*H*HAr), 3.63 (d, *J* = 14.8 Hz, 1H, CH*H*Ar) and 2.29 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 178.8 (C), 148.6 (C), 144.2 (C), 142.3 (C), 141.6 (C), 140.9 (C), 128.0 (C), 127.3 (CH), 126.3 (CH), 125.6 (CH), 125.4 (CH), 124.8 (CH), 124.1 (CH), 124.0 (CH), 123.3 (CH), 76.9 (CH), 73.7 (C), 66.8 (CH_2), 66.3 (CH), 38.4 (CH_2) and 24.9 (CH_2) ppm. IR (KBr) 3452 (O−H) and 1770 (C=O) cm^{-1}. MS (CI) *m*/*z* (%) 415 (MH^+). HRMS calcd for $C_{21}H_{19}O_5S_2$ (MH^+), 415.0674; found, 415.0674.

Sodium (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-1,4,5-trihydroxy-2-(thien-2-yl)methylcyclohex-2-en-1-carboxylate (9h). The experimental procedure was the same as for compound **5c** using carbolactone **19h** (20 mg), NaOH (97 μL), and THF (0.5 mL). Yield = 22 mg (99%). White solid; mp 184 °C (dec). $[\alpha]_D^{20} = -24°$ (*c* 1.5, H_2O). ^1H NMR (250 MHz, 50% CD_3OD/D_2O) δ 7.82 (m, 1H, ArH), 7.77 (m, 1H, ArH), 7.35 (m, 2H, 2 × ArH), 7.26 (s, 1H, ArH), 7.10 (dd, *J* = 3.3 and 3.0 Hz, 1H, ArH), 6.82 (d, *J* = 3.8 Hz, 2H, 2 × ArH), 5.25 (d, *J* = 12.0 Hz, 1H, OC*H*H), 5.04 (d, *J* = 12.0 Hz, 1H, OCH*H*), 4.36 (d, *J* = 5.2 Hz, 1H, H-4), 3.92 (m, 1H, H-5), 3.73 (d, *J* = 15.3 Hz, 1H, C*H*HAr), 3.31 (m, *J* = 15.3 Hz, 1H, CH*H*Ar) and 2.10 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, 50% CD_3OD/D_2O) δ 180.8 (C), 152.0 (C), 145.4 (C), 142.7 (C), 141.5 (C), 140.9 (C), 127.1 (CH), 126.3 (CH), 125.4 (CH), 125.3 (CH), 124.7 (CH), 123.7 (2 × CH + C), 123.3 (CH), 77.1 (C), 71.6 (CH), 70.0 (CH), 66.5 (CH_2), 37.4 (CH_2) and 27.2 (CH_2) ppm. IR (KBr) 3442 (O−H) and 1668 (C=O) cm^{-1}. MS (ESI) *m*/*z* (%) 431 (M^-). HRMS calcd for $C_{21}H_{19}O_6S_2$ (M^-), 431.0618; found, 431.0602.

(1*R*,4*S*,5*R*)-1,4-Di(*tert*-butyldimethylsilyloxy)-2-(5-methylbenzo[*b*]thiophen-2-yl)methyl-3-(thien-2-yl)methoxycyclohex-2-en-1,5-carbolactone (18i). A solution of ketone **12**[22] (350 mg,

0.87 mmol) in dry THF (17.5 mL), at room temperature and under argon, was treated with a solution of LHMDS (1.3 mL, 1.30 mmol, 1 M in THF). The resulting mixture was stirred for 1 h, and it was then treated with 2-iodomethyl-5-methylbenzo[*b*]thiophene[28] (480 mg, 1.75 mmol). After 40 min, water and diethyl ether were added, the organic layer was separated, and the aqueous phase was extracted with diethyl ether. The combined organic extracts were dried (anhyd Na_2SO_4), filtered, and concentrated under reduced pressure. The resulting residue was purified by flash chromatography, eluting with hexane and with diethyl ether/hexanes (10:90) to yield a diastereomeric mixture of *C*-alkyl products (158 mg, 33%). The resulting ketone was converted into compound **18i** by following the same experimental procedure as for compound **18g** using KHMDS (1.2 mL) in DMF (0.6 mL), 2-iodomethylthiophene (102 mg) in DMF (1.4 mL), and toluene (1.0 mL). Yield = 54 mg (29%). Yellow oil. $[\alpha]_D^{20} = -111°$ (*c* 1.3, $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.60 (d, *J* = 8.1 Hz, 1H, ArH), 7.42 (s, 1H, ArH), 7.27 (m, 1H, ArH), 7.05 (dd, *J* = 7.8 and 0.6 Hz, 1H, ArH), 6.92 (m, 3H, 3 × ArH), 4.97 (s, 2H, OCH_2), 4.56 (dd, *J* = 6.0 and 3.3 Hz, 1H, H-5), 4.43 (d, *J* = 3.3 Hz, 1H, H-4), 3.86 (d, *J* = 15.6 Hz, 1H, C*H*HAr), 3.74 (d, *J* = 15.6 Hz, 1H, CH*H*Ar), 2.56 (d, *J* = 10.8 Hz, 1H, H-6_{ax}), 2.44 (m, 4H, CH_3 + H-6_{eq}), 0.97 (s, 9H, $C(CH_3)_3$), 0.78 (s, 9H, $C(CH_3)_3$), 0.21 (s, 3H, $SiCH_3$), 0.20 (s, 3H, $SiCH_3$), 0.17 (s, 3H, $SiCH_3$) and 0.06 (s, 3H, $SiCH_3$) ppm. ^{13}C NMR (75 MHz, $CDCl_3$) δ 175.3 (C), 148.6 (C), 144.1 (C), 140.5 (C), 138.7 (C), 136.5 (C), 133.3 (C), 129.1 (C), 127.0 (CH), 126.7 (CH), 126.3 (CH), 124.8 (CH), 122.7 (CH), 121.5 (CH), 120.8 (CH), 77.4 (C), 74.7 (CH), 67.7 (CH_2), 67.3 (CH), 37.5 (2 × CH_2), 25.7 ($C(CH_3)_3$), 25.5 ($C(CH_3)_3$), 21.4 (CH_3), 18.1 ($C(CH_3)_3$), 18.0 ($C(CH_3)_3$), −3.3 ($SiCH_3$), −3.4 ($SiCH_3$) and −4.5 (2 × $SiCH_3$) ppm. IR (film) 1799 (C═O) cm^{-1}. MS (CI) *m*/*z* (%) 657 (MH^+).

(1*R*,4*S*,5*R*)-1,4-Dihydroxy-2-(5-methylbenzo[*b*]thiophen-2-yl)methyl-3-(thien-2-yl)methoxy cyclohex-2-en-1,5-carbolactone (19i). The experimental procedure was the same as for compound **15c** using silyl ether **18i** (40 mg), TBAF (0.14 μL), and THF (0.9 mL). Yield = 22 mg (88%). Yellow solid; mp 140−143 °C. $[\alpha]_D^{20} = -148°$ (*c* 1.2, CH_3OH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.56 (d, *J* = 8.2 Hz, 1H, ArH), 7.39 (s, 1H, ArH), 7.36 (d, *J* = 5.0 Hz, 1H, ArH), 7.03 (m, 2H, 2 × ArH), 6.94 (m, 2H, 2 × ArH), 6.97 (m, 2H, 2 × ArH), 5.25 (d, *J* = 12.2 Hz, 1H, OC*H*H), 5.15 (d, *J* = 12.2 Hz, 1H, OCH*H*), 4.63 (m, 1H, H-5), 4.54 (d, *J* = 3.2 Hz, 1H, H-4), 3.82 (d, *J* = 14.7 Hz, 1H, C*H*HAr), 3.69 (d, *J* = 14.7 Hz, 1H, CH*H*Ar) and 2.43−2.31 (m, 5H, CH_2-6 + CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 178.8 (C), 149.0 (C), 145.6 (C), 142.0 (C), 141.0 (C), 138.3 (C), 134.5 (C), 128.1 (CH), 127.7 (CH), 127.4 (CH), 125.9 (CH), 125.4 (C), 123.7 (CH), 122.5 (CH), 122.4 (CH), 76.9 (CH), 73.7 (C), 66.2 (CH_2), 65.9 (CH), 38.4 (CH_2), 25.9 (CH_2) and 21.5 (CH_2) ppm. IR (KBr) 3483 (O−H), 3429 (O−H) and 1729 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 541 (MNa^+). HRMS calcd for $C_{22}H_{20}O_5S_2Na$ (MNa^+), 451.0644; found, 451.0646.

Sodium (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-2-(5-methylbenzo[*b*]-thiophen-2-yl)methyl-3-(thien-2-yl)methoxycyclohex-2-en-1-carboxylate (9i). The experimental procedure was the same as for compound **5c** using carbolactone **19i** (20 mg), NaOH (100 μL), and THF (0.5 mL). Yield = 22 mg (97%). Beige solid; mp 191−194 °C. $[\alpha]_D^{20} = -55°$ (*c* 1.2, H_2O). ^{1}H NMR (250 MHz, D_2O) δ 7.59 (d, *J* = 8.0 Hz, 1H, ArH), 7.41 (s, 1H, ArH), 7.22 (d, *J* = 4.7 Hz, 1H, ArH), 7.02 (d, *J* = 8.0 Hz, 1H, ArH), 6.94 (s, 1H, ArH), 6.85 (m, 2H, 2 × ArH), 5.08 (d, *J* = 11.5 Hz, 1H, OC*H*H), 4.84 (d, *J* = 11.5 Hz, 1H, OCH*H*), 4.38 (d, *J* = 6.2 Hz, 1H, H-4), 3.98 (m, 1H, H-5), 3.68 (d, *J* = 16.0 Hz, 1H, C*H*HAr), 3.37 (d, *J* = 16.0 Hz, 1H, CH*H*Ar), 2.30 (s, 3H, CH_3), 2.21 (dd, *J* = 13.7 and 9.7 Hz, 1H, H-6_{ax}) and 2.09 (dd, *J* = 13.7 and 3.7 Hz, 1H, H-6_{eq}) ppm. ^{13}C NMR (63 MHz, D_2O) δ 180.3 (C), 151.7 (C), 146.0 (C), 140.9 (C), 139.5 (C), 137.0 (C), 134.8 (C), 128.5 (CH), 127.7 (CH), 127.5 (CH), 125.7 (CH), 123.3 (CH), 123.3 (C), 122.5 (CH), 122.0 (CH), 77.2 (C), 70.6 (CH), 70.3 (CH), 65.8 (OCH_2), 38.8 (CH_2), 28.3 (CH_2) and 21.1 (CH_3) ppm. IR (KBr) 3408 (O−H), 1660 (C═O) and 1605 (C═C) cm^{-1}. MS (ESI) *m*/*z* (%) 469 (MH^+). HRMS calcd for $C_{22}H_{22}O_6S_2Na$ (MH^+), 469.0750; found, 469.0747.

(1*R*,4*S*,5*R*)-1,4-Di(*tert*-butyldimethylsilyloxy)-2-(benzo[*b*]-thiophen-5-yl)methyl-3-(thien-2-yl)methoxycyclohex-2-en-1,5-carbolactone (18j). A solution of ketone **12**[22] (350 mg, 0.87 mmol) in dry THF (17.5 mL) was treated with LHMDS (1.3 mL, 1.31 mmol) at room temperature. After 1 h, 5-(iodomethyl)benzothiophene[28] (480 mg, 1.75 mmol) was added and the resultant mixture was stirred for 40 min. The reaction mixture was diluted with water and diethyl ether. The organic layer was separated, and the aqueous phase was extracted with diethyl ether (3 × 25 mL). The combined organic extracts were dried (anhyd Na_2SO_4), filtered, and concentrated. The residue was purified by flash chromatography on silica gel eluting with diethyl ether/hexanes [1) 0:100; 2) 10:90] to give the corresponding 2-alkyl ketone (158 mg, 33%). To a stirred solution of KHMDS (1.2 mL, 0.58 mmol, 0.5 M in toluene) in dry DMF (1.4 mL), under argon and at −78 °C, was added a solution of the previously obtained ketone (158 mg) in 2.8 mL of a mixture of dry DMF and dry toluene (1:1). The resultant solution was stirred at this temperature for 30 min. A solution of 2-(iodomethyl)thiophene (102 mg, 0.58 mmol) in a mixture of DMF and toluene (1.4:1, 2.4 mL), both dry, was then added. After 1 h, water and brine were added. The aqueous phase was extracted with diethyl ether (3 × 2 mL). The combined organic extracts were dried (anhyd $MgSO_4$), filtered, and concentrated. The residue was purified by flash chromatography on silica gel, eluting with diethyl ether/hexanes [(1) 5:95; (2) 10:90] to give compound **18j** (54 mg, 29%) as a colorless oil. ^{1}H NMR (250 MHz, $CDCl_3$) δ 7.55 (d, *J* = 8.2 Hz, 1H, ArH), 7.43 (s, 1H, ArH), 7.20 (d, *J* = 5.5 Hz, 1H, ArH), 7.11−6.97 (m, 5H, 5 × ArH), 4.73 (d, *J* = 12.2 Hz, 1H, OC*H*H), 4.68 (d, *J* = 12.2 Hz, 1H, OCH*H*), 4.39 (dd, *J* = 5.5 and 3.5 Hz, 1H, H-5), 4.26 (d, *J* = 3.5 Hz, 1H, H-4), 3.63 (d, *J* = 15.0 Hz, 1H, C*H*HAr), 3.49 (d, *J* = 15.0 Hz, 1H, CH*H*Ar), 2.37 (d, *J* = 11.0 Hz, 1H, H-6_{ax}), 2.25 (dd, *J* = 11.0 and 6.0 Hz, 1H, H-6_{eq}), 0.79 (s, 9H, $C(CH_3)_3$), 0.54 (s, 9H, $C(CH_3)_3$), 0.04 (s, 3H, $SiCH_3$), 0.02 (s, 3H, $SiCH_3$), −0.05 (s, 3H, $SiCH_3$) and −0.15 (s, 3H, $SiCH_3$) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ 175.5 (C), 148.3 (C), 139.7 (C), 138.8 (C), 136.0 (C), 130.3 (C), 127.8 (CH), 126.8 (CH), 126.6 (CH), 126.0 (CH), 125.4 (CH), 124.5 (C), 123.7 (CH), 123.1 (CH), 121.8 (CH), 75.0 (C), 74.7 (CH), 67.4 (CH_2), 67.3 (CH), 37.6 (CH_2), 30.1 (CH_2), 25.7 ($C(CH_3)_3$), 25.4 ($C(CH_3)_3$), 18.0 (2 × $C(CH_3)_3$), −3.3 ($SiCH_3$), −3.5 ($SiCH_3$) and −4.5 (2 × $SiCH_3$) ppm. MS (ESI) *m*/*z* (%) 643 (MH^+). HRMS calcd for $C_{33}H_{47}O_5S_2Si_2$ (MH^+), 643.2398; found, 643.2393.

(1*R*,4*S*,5*R*)-2-(Benzo[*b*]thiophen-5-yl)methyl-1,4-dihydroxy-3-(thien-2-yl)methoxycyclohex-2-en-1,5-carbolactone (19j). The experimental procedure was the same as for compound **15c** using silyl ether **18j** (40 mg), TBAF (140 μL), and THF (0.9 mL). Yield = 22 mg (88%). Yellow solid; mp 140−143 °C. ^{1}H NMR (250 MHz, CD_3OD) δ 7.67 (d, *J* = 8.2 Hz, 2H, 2 × ArH), 7.44 (d, *J* = 5.5 Hz, 1H, ArH), 7.36 (dd, *J* = 5.0 and 1.2 Hz, 1H, ArH), 7.26 (d, *J* = 8.2 Hz, 1H, ArH), 7.22 (d, *J* = 5.5 Hz, 1H, ArH), 6.97 (m, 2H, 2 × ArH), 5.21 (d, *J* = 12.2 Hz, 1H, OC*H*H), 5.09 (d, *J* = 12.2 Hz, 1H, OCH*H*), 4.62 (m, 1H, H-5), 4.55 (d, *J* = 3.5 Hz, 1H, H-4), 3.74 (d, *J* = 14.0 Hz, 1H, C*H*HAr), 3.57 (d, *J* = 14.0 Hz, 1H, CH*H*Ar), 2.38 (d, *J* = 11.0 Hz, 1H, H-6_{ax}) and 2.31 (dd, *J* = 11.0 and 5.2 Hz, 1H, H-6_{eq}) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 178.9 (C), 148.4 (C), 141.2 (C), 141.1 (C), 138.5 (C), 138.0 (C), 128.1 (CH), 127.7 (CH), 127.4 (CH), 127.1 (CH), 127.0 (CH + C), 124.8 (CH), 124.7 (CH), 122.6 (CH), 76.9 (CH), 73.9 (C), 66.2 (CH), 65.8 (CH_2), 38.5 (CH_2) and 30.6 (CH_2) ppm. MS (ESI) *m*/*z* (%) 437 (MNa^+). HRMS calcd for $C_{21}H_{18}O_5S_2Na$ (MNa^+), 437.0488; found, 437.0481.

Sodium (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(thien-2-yl)methoxy-2-(benzo[b]thiophen-5-yl)methylcyclohex-2-en-1-carboxylate (9j). The experimental procedure was the same as for compound

5c using carbolactone **19j** (20 mg), NaOH (97 μL), and THF (0.5 mL). Yield = 22 mg (97%). White solid; mp 199−201 °C. $[\alpha]_D^{20}$ = +57° (*c* 1.0, H_2O). ^{1}H NMR (250 MHz, 50% D_2O/CD_3CN) δ 8.27 (d, *J* = 8.5 Hz, 1H, ArH), 8.21 (br s, 1H, ArH), 8.01 (d, *J* = 5.5 Hz, 1H, ArH), 7.82 (m, 3H, 3 × ArH), 7.44 (m, 2H, 2 × ArH), 6.85 (m, 3H, 3 × ArH), 5.51 (d, *J* = 11.5 Hz, 1H, OC*H*H), 5.35 (d, *J* = 11.5 Hz, 1H, OCH*H*), 4.84 (m, 1H, H-4), 4.41 (m, 1H, H-5), 4.16 (d, *J* = 15.2 Hz, 1H, C*H*HAr), 3.66 (d, *J* = 15.7 Hz, 1H, CH*H*Ar) and 2.61 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, 50% D_2O/CD_3CN) δ 180.5 (C), 151.4 (C), 140.7 (C), 140.5 (C), 138.5 (C), 137.4 (C), 127.9 (CH), 127.6 (CH), 127.2 (CH), 127.2 (CH), 126.7 (CH), 124.8 (CH), 124.2 (CH), 123.7 (C), 122.4 (CH), 76.9 (C), 70.7 (CH), 69.5 (CH), 65.3 (CH_2), 37.2 (CH_2) and 32.4 (CH_2) ppm. IR (KBr) 3433 (O−H), 3290 (O−H) and 1687 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 455 (MH^+). HRMS calcd for $C_{21}H_{20}O_6S_2Na$ (MH^+), 455.0594; found, 455.0591.

Methyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (6a). A solution of the lactone **15a** (26 mg, 0.08 mmol) in dry methanol (0.9 mL) was treated with sodium methoxide (3.4 mg, 0.09 mmol). The resultant mixture was stirred at room temperature for 30 min, and the reaction mixture was diluted with ethyl acetate and water. The organic layer was separated, and the aqueous phase was extracted with ethyl acetate (×2). The combined organic extracts were dried (anhyd Na_2SO_4), filtered, and concentrated under reduced pressure. The obtained residue was purified by flash chromatography eluting with (50:50) ethyl acetate−hexanes to give methyl ester **6a** (15 mg, 53%) as a white solid; mp 152−154 °C. $[\alpha]_D^{20}$ = −40° (*c* 1.1, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.83 (m, 1H, ArH), 7.76 (m, 1H, ArH), 7.34 (m, 3H, 3 × ArH), 5.10 (s, 2H, OCH_2), 5.02 (s, 1H, H-2), 4.00 (m, 2H, H-4 + H-5), 3.66 (s, 3H, OMe) and 2.05 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 176.5 (C), 158.5 (C), 141.6 (C), 141.2 (C), 140.9 (C), 125.6 (CH), 125.4 (CH), 124.7 (CH), 124.5 (CH), 123.3 (CH), 99.9 (CH), 74.1 (CH), 73.9 (C), 70.4 (CH), 66.3 (CH_2), 53.1 (OCH_3), and 40.2 (CH_2) ppm. IR (KBr): 3435 (O−H) and 1726 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 373 (MNa^+). HRMS calcd for $C_{17}H_{18}O_6S_2Na$ (MNa^+), 373.0716; found, 373.0716.

Methyl (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(5-methylbenzo[*b*]-thiophen-2-yl)methoxycyclohex-2-ene-1-carboxylate (6b). The experimental procedure was the same as for compound **6a** using lactone **15b** (36 mg), NaOMe (6 mg), and MeOH (1.2 mL). Yield = 25 mg (62%). White solid; mp 183−185 °C. $[\alpha]_D^{20}$ = −31° (*c* 1.2, in MeOH). ^{1}H NMR (400 MHz, CD_3OD) δ 7.69 (d, *J* = 8.4 Hz, 1H, ArH), 7.56 (s, 1H, ArH), 7.28 (s, 1H, ArH), 7.16 (dd, *J* = 8.0 and 1.6 Hz, 1H, ArH), 5.07 (s, 2H, OCH_2Ar), 5.01 (s, 1H, H-2), 4.04−3.96 (m, 2H, H-4 + H-5), 3.67 (s, 3H, OCH_3), 2.43 (s, 3H, CH_3) and 2.05 (m, 2H, CH_2-6) ppm. ^{13}C NMR (100 MHz, CD_3OD) δ 176.5 (C), 158.5 (C), 141.2 (C), 141.2 (C), 138.8 (C), 135.3 (C), 127.3 (CH), 124.6 (CH), 124.3 (CH), 123.0 (CH), 99.9 (CH), 74.1 (CH), 73.9 (C), 70.5 (CH), 66.4 (CH_2), 53.1 (OCH_3), 40.2 (CH_2) and 21.4 (CH_3) ppm. IR (KBr) 3487 (O−H), 3435 (O−H) and 1718 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 387 (MNa^+). HRMS calcd for $C_{18}H_{20}O_6SNa$ (MNa^+), 387.0873; found, 387.0866.

Methyl (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(6-chlorobenzo[*b*]-thiophen-2-yl)methoxycyclohex-2-ene-1-carboxylate (6c). The experimental procedure was the same as for compound **6a** using lactone **15c** (16 mg), NaOMe (3 mg), and MeOH (0.4 mL). Yield = 7 mg (46%). Beige solid; mp 81−83 °C. $[\alpha]_D^{20}$ = −16° (*c* 1.0, in MeOH). ^{1}H NMR (250 MHz, acetone-d_6) δ 8.00 (d, *J* = 2.0 Hz, 1H, ArH), 7.82 (d, *J* = 8.5 Hz, 1H, ArH), 7.46 (s, 1H, ArH), 7.38 (dd, *J* = 8.5 and 2.0 Hz, 1H, ArH), 5.11 (d, *J* = 12.5 Hz, 1H, OC*H*HAr), 5.04 (d, *J* = 12.5 Hz, 1H, OCH*H*Ar), 5.00 (s, 1H, H-2), 4.51 (s, 1H, OH), 4.42 (s, 1H, OH), 4.20 (s, 1H, OH), 4.02 (m, 2H, H-4 + H-5), 3.66 (s, 3H, OCH_3) and 2.05 (m, 2H, CH_2-6) ppm. ^{13}C NMR (63 MHz, acetone-d_6) δ 177.0 (C), 159.0 (C), 143.3 (C), 140.1 (C), 131.8 (C), 126.9 (CH), 126.7 (CH), 124.6 (CH), 123.7 (CH + C), 100.8 (CH), 74.9 (CH), 74.4 (C), 71.2 (CH), 66.6 (CH_2), 53.7 (OCH_3) and 40.9 (CH_2) ppm. IR (KBr) 3436 (O−H) and 1739 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 407 (MNa^+). HRMS calcd for $C_{17}H_{17}O_6SClNa$ (MNa^+), 407.0327; found, 407.0330.

Methyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-yl)methoxy-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (6d). The experimental procedure was the same as for compound **6a** using lactone **15d** (52 mg), NaOMe (10 mg), and MeOH (1.8 mL). Yield = 38 mg (68%). White solid. $[\alpha]_D^{20}$ = −31° (*c* 1.4, MeOH). ^{1}H NMR (300 MHz, CD_3OD) δ 7.89 (d, *J* = 8.4 Hz, 2H, 2 × ArH), 7.58 (d, *J* = 5.7 Hz, 1H, ArH), 7.40 (dd, *J* = 8.4 and 1.2 Hz, 1H, ArH), 7.37 (d, *J* = 5.7 Hz, 1H, ArH), 4.98 (s, 1H, H-2), 4.94 (s, 2H, OCH_2), 4.02 (m, 2H, H-4 + H-5), 3.70 (s, 3H, OCH_3) and 2.04 (m, 2H, CH_2-6) ppm. ^{13}C NMR (75 MHz, CD_3OD) δ 176.7 (C), 158.9 (C), 141.3 (C), 134.3 (C), 128.2 (C), 128.1 (CH), 125.3 (CH), 124.9 (CH), 123.9 (CH), 123.4 (CH), 99.5 (CH), 74.3 (C), 74.0 (CH), 71.0 (OCH_2), 70.5 (CH), 53.1 (CH_3) and 40.3 (CH_2) ppm. IR (KBr) 3446 (O−H), 3305 (O−H) and 1732 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 373 (MNa^+). HRMS calcd for $C_{17}H_{18}O_6SNa$ (MNa^+), 373.0716; found, 373.0711. Elemental analysis $C_{17}H_{18}O_6S$ Calcd: C, 58.27; H, 5.18; S, 9.15. Found: C, 57.93; H, 5.36; S, 8.94.

Ethyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (7a). A solution of sodium carboxylate **5a** (6 mg, 0.02 mmol) and potassium carbonate (8 mg, 0.06 mmol) in DMF (0.3 mL) was stirred at room temperature for 30 min. Bromoethane (64 μL, 0.86 mmol) was added, and the reaction mixture was stirred for 2 h. Ethyl acetate was added, and the precipitate was filtered off. The resulting solution was washed with brine, dried (Na_2SO_4 anhyd), filtered, and concentrated under reduced pressure. The residue was purified by flash chromatography, eluting with ethyl acetate/hexanes (90:10) to give ester **7a** (3 mg, 49%) as a colorless oil. $[\alpha]_D^{20}$ = −13° (*c* 0.5, MeOH). ^{1}H NMR (500 MHz, CD_3OD) δ 7.83 (d, *J* = 7.5 Hz, 1H, ArH), 7.76 (dd, *J* = 7.0 and 1.5 Hz, 1H, ArH), 7.37 (s, 1H, ArH), 7.33 (m, 2H, 2 × ArH), 5.13 (s, 2H, OCH_2Ar), 5.01 (s, 1H, H-2), 4.10 (m, 2H, OCH_2CH_3), 3.99 (m, 2H, H-4 + H-5), 2.03 (m, 2H, CH_2-6) and 1.16 (t, *J* = 7.0 Hz, 3H, CH_3) ppm. ^{13}C NMR (125 MHz, CD_3OD) δ 176.1 (C), 158.4 (C), 141.6 (C), 141.4 (C), 140.9 (C), 125.6 (CH), 125.4 (CH), 124.7 (CH), 124.4 (CH), 123.3 (CH), 100.0 (CH), 74.2 (CH), 73.9 (C), 70.5 (CH), 66.3 (OCH_2), 62.7 (OCH_2), 40.3 (CH_2), and 14.4 (CH_3) ppm. IR (film): 3427 (O−H) and 1651 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 387 (MNa^+). HRMS calcd for $C_{18}H_{20}O_6SNa$ (MNa^+), 387.0873; found, 387.0870. Elemental analysis $C_{18}H_{20}O_6S$ Calcd: C, 59.33; H, 5.53; S, 8.80. Found: C, 58.96; H, 5.87; S, 8.81.

Ethyl (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(6-chlorobenzo[*b*]-thiophen-2-yl)methoxycyclohex-2-ene-1-carboxylate (7b). The experimental procedure was the same as for compound **7a** using compound **5c** (15 mg), potassium carbonate (16 mg), bromoethane (80 μL), and DMF (0.4 mL). Yield = 7 mg (46%). Colorless oil. $[\alpha]_D^{20}$ = −16° (*c* 1.0, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.88 (s, 1H, ArH), 7.73 (d, *J* = 8.5 Hz, 1H, ArH), 7.37 (s, 1H, ArH), 7.33 (dd, *J* = 8.5 and 2.0 Hz, 1H, ArH), 5.11 (s, 2H, OCH_2Ar), 5.00 (s, 1H, H-2), 4.11 (m, 2H, OCH_2CH_3), 3.99 (m, 2H, H-4 + H-5), 2.04 (m, 2H, CH_2-6) and 1.14 (t, *J* = 8.0 Hz, 3H, CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 176.0 (C), 158.4 (C), 142.7 (C), 142.5 (C), 139.5 (C), 131.6 (C), 126.2 (CH), 125.8 (CH), 123.9 (CH), 122.9 (CH), 100.1 (CH), 74.1 (C), 73.8 (CH), 70.5 (CH), 66.1 (OCH_2), 62.7 (OCH_2), 40.3 (CH_2) and 14.4 (CH_3) ppm. IR (KBr) 3419 (O−H), 1728 (C═O) and 1651 (C═C) cm^{-1}. MS (ESI) *mz*/*z* (%) 421 (MNa^+). HRMS calcd for $C_{18}H_{19}O_6SClNa$ (MNa^+), 421.0483; found, 421.0470.

Ethyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-yl)methoxyl-1,4,5-trihydroxycyclohex-2-ene-1-carboxylate (7c). The experimental procedure was the same as for compound **7a** using compound **5d** (63 mg), potassium carbonate (73 mg), bromoethane (400 μL), and

DMF (1.8 mL). Yield = 37 mg (58%). Yellow oil. $[\alpha]_D^{20} = -20°$ (*c* 1.1, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.88 (m, 2H, 2 × ArH), 7.57 (d, *J* = 5.5 Hz, 1H, ArH), 7.38 (m, 2H, 2 × ArH), 4.95 (br s, 3H, H-2 + OCH_2), 4.05 (m, 2H, OCH_2CH_3), 3.93 (m, 2H, H-4 + H-5), 2.05 (m, 2H, CH_2-6) and 1.19 (t, *J* = 7.2 Hz, 3H, OCH_2CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 176.2 (C), 158.8 (C), 141.3 (C), 140.7 (C), 134.3 (C), 128.1 (CH), 125.2 (CH), 124.9 (CH), 123.8 (CH), 123.4 (CH), 99.5 (CH), 74.3 (CH), 73.9 (C), 70.9 (OCH_2), 70.5 (CH), 62.7 (OCH_2), 40.3 (CH_2) and 14.4 (CH_3) ppm. IR (film) 3400 (O−H) and 1728 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 387 (MNa^+). HRMS calcd for $C_{18}H_{20}O_6SNa$ (MNa^+), 387.0873; found, 387.0870.

Ethyl (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(naphth-2-yl)methoxycyclohex-2-enecarboxylate (7d). The experimental procedure was the same as for compound **7a** using compound **5e** (7 mg), potassium carbonate (8 mg), bromoethane (40 μL), and DMF (0.2 mL). Yield: 3 mg (42%). Yellow oil. $[\alpha]_D^{20} = -39°$ (*c* 1.2, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.88 (m, 4H, 4 × ArH), 7.51 (m, 3H, 3 × ArH), 5.01 (s, 2H, CH_2O), 4.98 (s, 1H, H-2), 4.08 (m, 4H, OCH_2CH_3 + H-4 + H-5), 2.04 (m, 2H, CH_2-6) and 1.17 (t, *J* = 7.2 Hz, 3H, CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 176.2 (C), 158.9 (C), 135.7 (C), 134.8 (C), 134.6 (C), 129.2 (CH), 129.0 (CH), 128.7 (CH), 127.5 (CH), 127.3 (CH), 127.1 (CH), 126.6 (CH), 99.6 (CH), 74.3 (CH), 73.9 (C), 70.9 (OCH_2), 70.5 (CH), 62.6 (CH_2), 40.3 (CH_2) and 14.4 (CH_3) ppm. IR (film): 3419 (O−H) and 1649 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 381 (MNa^+). HRMS calcd for $C_{20}H_{22}O_6Na$ (MNa^+), 381.1309; found, 381.1307.

Propyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-2-yl)methoxy-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (8a). The experimental procedure was the same as for compound **7a** using compound **5a** (15 mg), potassium carbonate (17 mg), 1-bromopropane (0.1 mL), and DMF (0.4 mL). Yield = 6 mg (38%). Colorless oil. $[\alpha]_D^{20} = -27°$ (*c* 1.0, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.83 (m, 1H, ArH), 7.76 (m, 1H, ArH), 7.33 (m, 3H, 3 × ArH), 5.13 (s, 2H, OCH_2Ar), 5.00 (s, 1H, H-2), 4.00 (m, 4H, $OCH_2CH_2CH_3$ + H-4 + H-5), 2.04 (m, 2H, CH_2-6), 1.53 (m, 2H, $OCH_2CH_2CH_3$) and 0.85 (t, *J* = 7.2 Hz, 3H, CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 176.2 (C), 158.4 (C), 141.6 (C), 141.5 (C), 140.9 (C), 125.6 (CH), 125.4 (CH), 124.7 (CH), 124.3 (CH), 123.3 (CH), 100.1 (CH), 74.2 (CH), 73.9 (C), 70.5 (CH), 68.2 (OCH_2), 66.3 (OCH_2), 40.4 (CH_2), 22.9 (CH_2) and 10.6 (CH_3) ppm. IR (film): 3375 (O−H) and 1730 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 401 (MNa^+). HRMS calcd for $C_{19}H_{22}O_6Na$ (MNa^+), 401.1029; found, 401.1030.

Propyl (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(6-chlorobenzo[*b*]-thiophen-2-yl)methoxycyclohex-2-ene-1-carboxylate (8b). The experimental procedure was the same as for compound **7a** using compound **5c** (15 mg), potassium carbonate (16 mg), 1-bromopropane (0.1 mL), and DMF (0.4 mL). Yield = 6 mg (38%). Yellow oil. ^{1}H NMR (250 MHz, CD_3OD) δ 7.88 (d, *J* = 1.8 Hz, 1H, ArH), 7.73 (d, *J* = 8.5 Hz, 1H, ArH), 7.36 (s, 1H, ArH), 7.33 (dd, *J* = 8.5 and 2.0 Hz, 1H, ArH), 5.12 (s, 2H, OCH_2Ar), 4.99 (s, 1H, H-2), 4.11 (m, 2H, OCH_2Ar), 3.99 (m, 4H, OCH_2 + H-4 + H-5), 2.03 (m, 2H, CH_2-6), 1.53 (q, *J* = 6.7 Hz, 2H, CH_2CH_3) and 0.85 (t, *J* = 7.2 Hz, 3H, CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 176.1 (C), 158.3 (C), 142.7 (C), 142.6 (C), 139.5 (C), 131.6 (C), 126.2 (CH), 125.8 (CH), 123.8 (CH), 122.9 (CH), 100.2 (CH), 74.2 (C), 73.9 (CH), 70.5 (CH), 68.2 (OCH_2), 66.1 (OCH_2), 40.4 (CH_2), 22.9 (CH_2) and 10.6 (CH_3) ppm. IR (film): 3421 (O−H) and 1720 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 435 (MNa^+). HRMS calcd for $C_{19}H_{21}O_6ClNa$ (MNa^+), 435.0640; found, 435.0638.

Propyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-yl)methoxy-1,4,5-trihydroxycyclohex-2-ene-1-carboxylate (8c). The experimental procedure was the same as for compound **7a** using compound **5d** (12 mg), potassium carbonate (14 mg), 1-bromopropane (85 μL), and DMF (0.3 mL). Yield = 4 mg (32%). Colorless oil. $[\alpha]_D^{20} = -46°$ (*c* 1.2, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ δ 7.86 (m, 2H, 2 × ArH), 7.55 (d, *J* = 5.6 Hz, 1H, ArH), 7.37 (dd, *J* = 8.8 and 1.2 Hz, 1H, ArH), 7.34 (d, *J* = 5.6 Hz, 1H, ArH), 4.99 (d, *J* = 11.6 Hz, 1H, OC*H*HAr), 4.94 (m, 2H, OCH*H*Ar + H-2), 4.02 (m, 4H, $OCH_2CH_2CH_3$ + H-4 + H-5), 2.04 (m, 2H, CH_2-6), 1.58 (m, 2H, $OCH_2CH_2CH_3$) and 0.89 (t, *J* = 7.2 Hz, 3H, $OCH_2CH_2CH_3$) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 176.2 (C), 158.8 (C), 141.3 (C), 140.7 (C), 134.3 (C), 128.1 (CH), 125.2 (CH), 124.9 (CH), 123.8 (CH), 123.4 (CH), 99.5 (CH), 74.3 (CH), 73.9 (C), 70.9 (OCH_2), 70.5 (CH), 62.7 (OCH_2), 40.3 (CH_2) and 14.4 (CH_3) ppm. IR (film): 3425 (O−H) and 1643 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 401 (MNa^+). HRMS calcd for $C_{19}H_{22}O_6SNa$ (MNa^+), 401.1029; found, 401.1020.

Propyl (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(naphth-2-yl)methoxycyclohex-2-ene-1-carboxylate (8d). The experimental procedure was the same as for compound **7a** using compound **5e** (30 mg), potassium carbonate (35 mg), 1-bromopropane (0.22 mL), and DMF (0.8 mL). Yield = 16 mg (50%). Orange oil. $[\alpha]_D^{20} = -36°$ (*c* 1.0, MeOH). ^{1}H NMR (300 MHz, CD_3OD) δ 7.86 (m, 4H, 4 × ArH), 7.52 (dd, *J* = 8.4 and 1.5 Hz, 1H, ArH), 7.49−7.46 (m, 2H, 2 × ArH), 5.04 (d, *J* = 12.0 Hz, 1H, C*H*HO), 4.98 (d, *J* = 12.0 Hz, 1H, CH*H*O), 4.97 (s, 1H, H-2), 4.21 (dd, *J* = 5.4 and 0.9 Hz, 1H, H-5), 4.01 (m, 3H, $OCH_2CH_2CH_3$ + H-4), 2.06 (m, 2H, CH_2-6), 1.54 (m, 2H, $OCH_2CH_2CH_3$) and 0.86 (t, *J* = 7.5 Hz, 3H, $OCH_2CH_2CH_3$) ppm. ^{13}C NMR (75 MHz, CD_3OD) δ 176.2 (C), 158.9 (C), 135.7 (C), 134.8 (C), 134.6 (C), 129.2 (CH), 129.0 (CH), 128.7 (CH), 127.4 (CH), 127.2 (CH), 127.1 (CH), 126.5 (CH), 99.7 (CH), 74.4 (CH), 74.0 (C), 70.9 (OCH_2), 70.5 (CH), 68.1 (OCH_2), 40.4 (CH_2), 22.9 (CH_2) and 10.6 (CH_3) ppm. IR (film) 3377 (OH) and 1730 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 395 (MNa^+). HRMS calcd for $C_{21}H_{24}O_6Na$ (MNa^+), 395.1465; found, 395.1461.

Methyl (1*R*,4*S*,5*R*)-3-(5-Methylbenzo[*b*]thiophen-2-yl)-methoxy-2-(5-methylbenzo[*b*]thiophen-2-yl)methyl-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (10a). The experimental procedure was the same as for compound **2a** using carbolactone **16b** (123 mg), NaOMe (15 mg), and MeOH (2.8 mL). Yield = 32 mg (24%). Beige solid; mp 168−170 °C. $[\alpha]_D^{20} = +48°$ (*c* 1.2, MeOH). ^{1}H NMR (250 MHz, acetone-d_6) δ 7.72 (d, *J* = 8.2 Hz, 1H, ArH), 7.60 (d, *J* = 8.2 Hz, 1H, ArH), 7.53 (s, 1H, ArH), 7.31 (s, 1H, ArH), 7.22 (s, 1H, ArH), 7.16 (dd, *J* = 8.2 and 1.2 Hz, 1H, ArH), 7.05 (dd, *J* = 8.2 and 1.2 Hz, 1H, ArH), 6.83 (s, 1H, ArH), 5.51 (d, *J* = 12.2 Hz, 1H, OC*H*HAr), 5.27 (d, *J* = 12.2 Hz, 1H, OCH*H*Ar), 4.60 (d, *J* = 6.5 Hz, 1H, OH), 4.49 (s, 1H, OH), 4.38 (t, *J* = 6.5 Hz, 1H, H-4), 4.05 (m, 1H, H-5), 3.62 (s, 2H, CH_2Ar), 3.27 (s, 3H, OMe), 2.41 (s, 3H, Me), 2.36 (s, 3H, Me), 2.16 (dd, *J* = 13.0 and 11.3 Hz, 1H, H-6_{ax}) and 1.99 (m, 1H, H-6_{eq}) ppm. ^{13}C NMR (63 MHz, acetone-d_6) δ 177.1 (C), 154.7 (C), 146.6 (C), 143.4 (C), 142.2 (C), 141.7 (C), 139.3 (C), 138.7 (C), 135.6 (C), 135.1 (C), 127.9 (CH), 126.7 (CH), 125.4 (CH), 124.7 (CH), 124.4 (CH), 123.8 (CH), 123.3 (CH), 123.2 (CH), 120.1 (C), 77.0 (C), 73.6 (CH), 71.9 (CH), 67.9 (OCH_2), 53.6 (OCH_3), 42.1 (CH_2), 29.0 (CH_3), 22.4 (CH_3) and 22.3 (CH_3) ppm. IR (KBr): 3502 (O−H), 3435 (O−H) and 1730 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 547 (MNa^+). HRMS calcd for $C_{28}H_{28}O_6S_2Na$ (MNa^+), 547.1220; found, 547.1215. Elemental analysis $C_{28}H_{28}O_6S_2 \cdot H_2O$ Calcd: C, 61.97; H, 5.57; S, 11.82. Found: C, 61.40; H, 5.30; S, 11.52.

Methyl (1*R*,4*S*,5*R*)-3-(6-Chlorobenzo[*b*]thiophen-5-yl)-methoxy-2-(6-chlorobenzo[*b*]thiophen-5-yl)methyl-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (10b). The experimental procedure was the same as for compound **2a** using lactone **16c** (42 mg), NaOMe (5 mg), and MeOH (0.9 mL). Yield = 22 mg (49%). Beige solid; mp 154−156 °C. $[\alpha]_D^{20} = +74°$ (*c* 1.1, MeOH). ^{1}H NMR (300 MHz, acetone-d_6) δ 7.90 (d, *J* = 1.8 Hz, 1H, ArH), 7.77 (d, *J* = 2.1 Hz, 1H, ArH), 7.74 (d, *J* = 8.4 Hz, 1H, ArH), 7.55 (d, *J* = 8.4 Hz, 1H, ArH), 7.34 (dd, *J* = 8.4 and 1.8 Hz, 1H, ArH), 7.31 (d, *J* = 0.9 Hz, 1H, ArH), 7.24 (dd, *J* = 8.4 and 2.1 Hz, 1H, ArH), 6.97 (d, *J* = 0.9 Hz, 1H, ArH), 5.52 (dd, *J* = 12.6 and 0.9 Hz, 1H, OC*H*HAr), 5.27 (dd, *J* = 12.6 and 0.9 Hz, 1H, OCH*H*Ar), 4.69 (d, *J* = 6.9 Hz, 1H, OH), 4.58 (s, 1H, OH), 4.55 (d, *J* = 3.9 Hz, 1H, OH), 4.38 (t, *J* = 6.6 Hz, 1H, H-4), 4.05 (m, 1H, H-5), 3.66 (d, *J* = 15.6 Hz, 1H, C*H*HAr), 3.59 (d, *J* = 15.6 Hz, 1H, CH*H*Ar), 3.34 (s, 3H, OMe), 2.16

(dd, J = 13.2 and 11.1 Hz, 1H, H-6_{ax}) and 2.01 (m, 1H, H-6_{eq}) ppm. ^{13}C NMR (75 MHz, acetone-d_6) δ 177.1 (C), 154.8 (C), 147.9 (C), 144.4 (C), 143.3 (C), 142.8 (C), 140.5 (C), 140.0 (C), 131.7 (C), 130.5 (C), 126.7 (CH), 126.6 (CH), 126.2 (CH), 125.5 (CH), 124.3 (CH), 123.6 (CH), 123.1 (CH), 123.0 (CH), 119.9 (C), 77.1 (C), 73.4 (CH), 71.8 (CH), 67.7 (OCH_2), 53.7 (OCH_3), 42.1 (CH_2) and 29.2 (CH_2) ppm. IR (KBr) 3419 (O−H) and 1730 (C═O) cm^{-1}. MS (ESI) m/z (%) 587 (MNa^+). HRMS calcd for $C_{26}H_{22}O_6S_2Cl_2Na$ (MNa^+), 587.0127; found, 587.0123. Elemental analysis $C_{26}H_{22}O_6S_2Cl_2.H_2O$ Calcd: C, 53.52; H, 4.15; S, 10.99. Found: C, 53.72; H, 4.36; S, 11.34.

Methyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-yl)methoxy-2-(benzo[*b*]thiophen-5-yl)methyl-1,4,5-trihydroxycyclohex-2-enecarboxylate (10c). A solution of carbolactone **16d** (57 mg, 0.12 mmol) in dry methanol (0.7 mL) and acetonitrile (0.7 mL) was treated with sodium methoxide (7 mg, 0.13 mmol). The resultant mixture was stirred at room temperature for 2 h and was diluted with ethyl acetate and water. The organic layer was separated, and the aqueous phase was extracted with ethyl acetate (×3). The combined organic extracts were filtered (anhyd Na_2SO_4) and concentrated under reduced pressure. The residue was purified by flash chromatography on silica gel, eluting with (1:1) ethyl acetate−hexanes to afford methyl ester **10c** (24 mg, 40%) as a white solid; mp 148−152 °C. $[\alpha]_D^{20}$ = +9° (c 1.2, MeOH). ^{1}H NMR (300 MHz, CD_3OD) δ 7.72 (d, J = 8.4 Hz, 1H, ArH), 7.60 (m, 2H, 2 × ArH), 7.52 (br s, 1H, ArH), 7.48 (d, J = 5.4 Hz, 1H, ArH), 7.42 (d, J = 5.4 Hz, 1H, ArH), 7.23 (dd, J = 8.1 and 1.5 Hz, 1H, ArH), 7.17 (dd, J = 5.7 and 0.6 Hz, 1H, ArH), 7.11 (m, 2H, 2 × ArH), 5.26 (d, J = 11.1 Hz, 1H, OC*H*HAr), 4.87 (d, J = 11.1 Hz, 1H, OCH*H*Ar), 4.38 (d, J = 7.5 Hz, 1H, H-4), 4.03 (m, 1H, H-5), 3.55 (d, 1H, J = 15.3 Hz, C*H*HAr), 3.38 (d, 1H, J = 15.3 Hz, CH*H*Ar), 3.14 (s, 3H, OCH_3), 2.18 (dd, J = 13.2 and 12.0 Hz, 1H, H-6_{ax}) and 1.99 (dd, J = 13.2 and 3.9 Hz, 1H, H-6_{eq}) ppm. ^{13}C NMR (75 MHz, CD_3OD) δ 176.6 (C), 154.0 (C), 141.2 (2 × C), 140.6 (C), 138.5 (C), 137.5 (C), 135.1 (C), 127.9 (CH), 127.2 (CH), 127.0 (CH), 125.9 (CH), 124.9 (CH), 124.7 (2 × CH), 124.4 (CH), 123.2 (CH), 122.5 (CH), 120.8 (C), 77.0 (C), 72.9 (CH), 72.2 (CH_2), 71.3 (CH), 52.7 (OCH_3), 41.5 (CH_2) and 33.1 (CH_2) ppm. IR (KBr) 3410 (O−H) and 1734 (C═O) cm^{-1}. MS (ESI) m/z (%) 519 (MNa^+). HRMS calcd for $C_{26}H_{24}O_6S_2Na$ (MNa^+), 519.0907; found, 519.0901. Elemental analysis $C_{26}H_{24}O_6S_2 \cdot 1/2H_2O$ Calcd: C, 61.76; H, 4.98; S, 12.68. Found: C, 61.47; H, 5.28; S, 12.30.

Methyl (1*R*,4*S*,5*R*)-1,4-Dihydroxy-3-(thien-2-yl)methoxy-2-(thien-2-yl)methylcyclohex-2-en-1-carboxylate (10d). The experimental procedure was the same as for compound **2a** using lactone **16f** (40 mg), NaOMe (6 mg), and MeOH (1.2 mL). Yield = 22 mg (51%). Beige solid; mp 55−57 °C. $[\alpha]_D^{20}$ = +7° (c 1.1, MeOH). ^{1}H NMR (300 MHz, CD_3OD) δ 7.34 (dd, J = 5.1 and 1.2 Hz, 1H, ArH), 7.10 (dd, J = 5.1 and 1.2 Hz, 1H, ArH), 7.04 (m, 1H, ArH), 6.95 (dd, J = 5.1 and 3.3 Hz, 1H, ArH), 6.80 (dd, J = 5.1 and 3.3 Hz, 1H, ArH), 6.67 (m, 1H, ArH), 5.37 (d, J = 11.4 Hz, 1H, OC*H*HAr), 5.01 (d, J = 11.4 Hz, 1H, OCH*H*Ar), 4.25 (d, J = 7.8 Hz, 1H, H-4), 3.93 (m, 1H, H-5), 3.61 (d, J = 15.3 Hz, 1H, C*H*HAr), 3.43 (d, J = 15.3 Hz, 1H, CH*H*Ar), 3.33 (s, 3H, OCH_3), 2.14 (dd, J = 12.9 and 12.0 Hz, 1H, H-6_{ax}) and 1.95 (dd, J = 12.9 and 3.9 Hz, 1H, H-6_{eq}) ppm. ^{13}C NMR (75 MHz, CD_3OD) δ 176.2 (C), 153.7 (C), 144.1 (C), 141.1 (C), 128.1 (CH), 127.4 (CH), 127.2 (CH), 126.9 (CH), 126.4 (CH), 124.3 (CH), 120.9 (C), 76.3 (C), 72.7 (CH), 71.1 (CH), 66.5 (CH_2), 52.8 (CH_3), 41.3 (CH_2) and 27.2 (CH_2) ppm. IR (KBr) 3435 (O−H) and 1734 (C═O) cm^{-1}. MS (ESI) m/z (%) 419 (MNa^+). HRMS calcd for $C_{18}H_{20}O_6S_2Na$ (MNa^+), 419.0594; found, 419.0581. Elemental analysis $C_{18}H_{20}O_6S_2 \cdot 1/4H_2O$ Calcd; C, 53.92; H, 5.15; S, 15.99. Found: C, 54.15; H, 5.46; S, 15.65.

Ethyl (1*R*,4*S*,5*R*)-3-(6-Chlorobenzo[*b*]thiophen-5-yl)-methoxy-2-(6-chlorobenzo[*b*]thiophen-5-yl)methyl-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (11a). The experimental procedure was the same as for compound **7a** using compound **9c** (33 mg), potassium carbonate (24 mg), bromoethane (120 μL), and DMF (0.6 mL). Yield = 7 mg (21%). Yellow oil. $[\alpha]_D^{20}$ = +32° (c 0.7, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.77 (d, J = 2.0 Hz, 1H, ArH), 7.65 (m, 2H, 2 × ArH), 7.39 (d, J = 8.5 Hz, 1H, ArH), 7.30 (dd, J = 8.5 and 2.0 Hz, 1H, ArH), 7.18 (m, 2H, 2 × ArH), 6.84 (s, 1H, ArH), 5.42 (d, J = 12.5 Hz, 1H, OC*H*HAr), 5.22 (d, J = 12.5 Hz, 1H, OCH*H*Ar), 4.33 (d, J = 7.5 Hz, 1H, H-4), 3.99 (m, 1H, H-5), 3.79 (m, 2H, CH_2CH$_3$), 3.88−3.58 (m, 4H, OCH_2CH$_3$ + CH_2Ar), 2.18 (t, J = 13.0 Hz, 1H, H-6_{ax}), 1.98 (dd, J = 13.0 and 4.0 Hz, 1H, H-6_{eq}) and 0.89 (t, J = 7.2 Hz, 3H, CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 175.9 (C), 154.1 (C), 146.7 (C), 143.3 (C), 143.0 (C), 142.2 (C), 139.9 (C), 139.4 (C), 131.5 (C), 130.3 (C), 126.1 (CH), 125.8 (CH), 125.6 (CH), 124.6 (CH), 124.1 (CH), 122.9 (CH), 122.4 (CH), 122.3 (CH), 119.9 (C), 76.6 (C), 72.7 (CH), 71.2 (CH), 67.0 (OCH_2), 62.9 (CH_2), 41.4 (CH_2), 28.5 (CH_2) and 14.0 (CH_3) ppm. IR (film) 3419 (O−H) and 1643 (C═O) cm^{-1}. MS (ESI) m/z (%) 601 (MNa^+). HRMS calcd for $C_{27}H_{24}O_6S_2Cl_2Na$ (MNa^+), 601.0284; found, 601.0283.

Ethyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-yl)methoxy-2-(benzo[*b*]thiophen-5-yl)methyl-1,4,5-trihydroxycyclohex-2-en-1-carboxylate (11b). The experimental procedure was the same as for compound **7a** using compound **9d** (8 mg), potassium carbonate (6 mg), bromoethane (33 μL), and DMF (0.2 mL). Yield = 4 mg (49%). Colorless oil. $[\alpha]_D^{20}$ = +4° (c 0.5, MeOH). ^{1}H NMR (400 MHz, CD_3OD) δ 7.77 (d, J = 8.4 Hz, 1H, ArH), 7.63 (m, 2H, 2 × ArH), 7.56 (s, 1H, ArH), 7.53 (d, J = 5.2 Hz, 1H, ArH), 7.45 (d, J = 5.2 Hz, 1H, ArH), 7.26 (d, J = 8.4 Hz, 1H, ArH), 7.21 (d, J = 5.2 Hz, 1H, ArH), 7.15 (m, 2H, 2 × ArH), 5.27 (d, J = 11.2 Hz, 1H, OC*H*HAr), 4.91 (d, J = 11.2 Hz, 1H, OCH*H*Ar), 4.37 (d, J = 7.6 Hz, 1H, H-4), 4.01 (m, 1H, H-5), 3.77 (m, 2H, OCH_2CH$_3$), 3.41 (m, 2H, CH_2Ar), 2.17 (t, J = 13.2 Hz, 1H, H-6_{ax}), 1.97 (dd, J = 13.2 and 4.0 Hz, 1H, H-6_{eq}) and 0.91 (t, J = 7.2 Hz, 3H, CH_3) ppm. ^{13}C NMR (100 MHz, CD_3OD) δ 176.2 (C), 154.0 (C), 141.2 (C), 141.1 (C), 138.5 (C), 137.6 (C), 135.2 (C), 127.9 (CH), 127.2 (CH), 127.1 (CH), 126.0 (CH), 125.0 (CH), 124.7 (CH), 124.7 (CH), 124.5 (CH), 124.5 (C), 123.2 (CH), 122.5 (CH), 120.9 (C), 77.0 (C), 73.0 (CH), 72.2 (OCH_2), 71.4 (CH), 62.7 (CH_2), 41.6 (CH_2), 33.2 (CH_2) and 14.0 (CH_3) ppm. IR (film) 3421 (O−H) and 1639 (C═O) cm^{-1}. MS (ESI) m/z (%) 533 (MNa^+). HRMS calcd for $C_{27}H_{26}O_6S_2Na$ (MNa^+), 533.1063; found, 533.1058.

Ethyl (1*R*,4*S*,5*R*)-1,4,5-Trihydroxy-3-(thien-2-yl)methoxy-2-(5-methylbenzo[*b*]thiophen-2-yl)methylcyclohex-2-en-1-carboxylate (11c). The experimental procedure was the same as for compound **7a** using compound **9i** (33 mg), potassium carbonate (15 mg), bromoethane (80 μL), and DMF (0.4 mL). Yield = 6 mg (37%). Yellow oil. $[\alpha]_D^{20}$ = +24° (c 0.8, MeOH). ^{1}H NMR (400 MHz, CD_3OD) δ 7.54 (d, J = 8.4 Hz, 1H, ArH), 7.37 (s, 1H, ArH), 7.31 (dd, J = 4.8 and 0.8 Hz, 1H, ArH), 7.01 (m, 2H, 2 × ArH), 6.91 (dd, J = 5.2 and 3.6 Hz, 1H, ArH), 6.78 (s, 1H, ArH), 5.36 (d, J = 11.6 Hz, 1H, OC*H*HAr), 5.08 (d, J = 11.6 Hz, 1H, OCH*H*Ar), 4.26 (d, J = 7.6 Hz, 1H, H-4), 3.94 (m, 1H, H-5), 3.83 (m, 1H, CH_3CH*H*O), 3.62 (d, J = 15.2 Hz, 1H, C*H*HAr), 3.48 (d, J = 15.2 Hz, 1H, CH*H*Ar), 3.78 (m, 1H, CH_3C*H*HO), 2.37 (s, 3H, CH_3), 2.14 (dd, J = 12.4 and 12.8 Hz, 1H, H-6_{ax}), 1.94 (dd, J = 12.8 and 4.0 Hz, 1H, H-6_{eq}) and 0.89 (t, J = 7.2 Hz, 3H, CH_3CH$_2$O) ppm. ^{13}C NMR (100 MHz, CD_3OD) δ 176.0 (C), 154.1 (C), 145.6 (C), 141.7 (C), 141.2 (C), 138.4 (C), 134.7 (C), 128.3 (CH), 127.6 (CH), 127.4 (CH), 126.1 (CH), 123.7 (CH), 122.7 (CH), 122.5 (CH), 120.2 (C), 76.5 (C), 72.8 (CH), 71.2 (CH), 66.5 (OCH_2), 62.9 (OCH_2), 41.5 (CH_2), 28.3 (CH_2), 21.5 (CH_3) and 14.0 (CH_3) ppm. IR (film) 3435 (O−H), 1722 (C═O) and 1651 (C═C) cm^{-1}. MS (ESI) m/z (%) 497 (MNa^+). HRMS calcd for $C_{24}H_{26}O_6S_2Na$ (MNa^+), 497.1063; found, 497.1054.

Ethyl (1*R*,4*S*,5*R*)-4,5-Dibutyroxy-3-(benzo[*b*]thiophen-5-yl)methoxy-1-hydroxycyclohex-2-enecarboxylate (7e). To a stirred solution of ethyl ester **7c** (22 mg, 0.06 mmol) and pyridine (17 μL, 0.21 mmol) in dry acetonitrile (1.5 mL) at 0 °C was added

butyryl chloride (22 μL, 0.21 mmol). The resulting solution was stirred at room temperature for 10 h, and it was then partitioned into water and ethyl acetate. The organic layer was separated, and the aqueous phase was extracted with ethyl acetate (×3). The combined organic extracts were dried (anhyd $MgSO_4$), filtered, and concentrated under reduced pressure. The residue was purified by flash chromatography eluting with (50:50) ethyl acetate/hexane to afford ester **7e** (11 mg, 36%) as a yellow oil. $[\alpha]_D^{20} = -26°$ (*c* 1.0, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.88 (d, *J* = 8.5 Hz, 1H, ArH), 7.81 (br s, 1H, ArH), 7.59 (d, *J* = 5.5 Hz, 1H, ArH), 7.35 (d, *J* = 5.5 Hz, 1H, ArH), 7.31 (br d, *J* = 8.5 Hz, 1H, ArH), 5.63 (d, *J* = 7.8 Hz, 1H, H-4), 5.44 (m, 1H, H-5), 5.18 (s, 1H, H-2), 4.94 (s, 2H, OCH_2Ar), 4.16 (m, 2H, OCH_2CH_3), 2.26 (m, 2H, CH_2-6), 1.70−1.48 (m, 4H, 2 × CH_2), 1.36−1.11 (m, 4H, 2 × CH_2), 0.97 (m, 6H, 2 × CH_3) and 0.79 (t, *J* = 7.2 Hz, 3H, OCH_2CH_3) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 175.1 (C), 174.7 (C), 174.1 (C), 154.8 (C), 141.3 (C), 140.8 (C), 134.0 (C), 128.3 (CH), 125.0 (CH), 124.9 (CH), 123.6 (CH), 123.5 (CH), 101.8 (CH), 73.4 (C), 71.6 (CH), 71.1 (OCH_2), 70.6 (CH), 62.9 (OCH_2), 37.4 (CH_2), 37.0 (CH_2), 37.0 (CH_2), 19.5 (CH_2), 19.4 (CH_2), 14.4 (CH_3), 13.9 (CH_3) and 13.9 (CH_3) ppm. IR (film) 3398 (O−H) and 1736 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 527 (MNa^+). HRMS calcd for $C_{26}H_{32}O_8SNa$ (MNa^+), 527.1710; found, 527.1695.

Propyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-ylmethoxy)-4,5-dibutyroxy-1-hydroxycyclohex-2-enecarboxylate (8e). A stirred solution of ethyl ester **8c** (3.4 mg, 8.99 μmol) in dry acetonitrile (0.2 mL) at 0 °C was treated with a solution of dry pyridine (30 μL, 1 M in acetonitrile, 0.03 mmol). After 10 min, a solution of butyryl chloride (30 μL, 1 M in acetonitrile, 0.03 mmol) was added. The resuting solution was stirred at room temperature for 10 h and then partitioned into water and ethyl acetate. The organic layer was separated, and the aqueous phase was extracted with ethyl acetate (×3). The combined organic extracts were dried (anhyd $MgSO_4$), filtered, and concentrated under reduced pressure. The residue was purufied by flash chromatography on silica gel, eluting with ethyl acetate/hexane (35:65) to give ester **8e** (3 mg, 68%) as a yellow oil. ^{1}H NMR (500 MHz, CD_3OD) δ 7.88 (d, *J* = 8.0 Hz, 1H, ArH), 7.80 (s, 1H, ArH), 7.59 (d, *J* = 5.5 Hz, 1H, ArH), 7.36 (d, *J* = 5.5 Hz, 1H, ArH), 7.31 (br d, *J* = 8.0 Hz, 1H, ArH), 5.63 (d, *J* = 8.0 Hz, 1H, H-4), 5.45 (m, 1H, H-5), 5.18 (s, 1H, H-2), 4.94 (s, 2H, OCH_2Ar), 4.06 (t, *J* = 6.5 Hz, 2H, $OCH_2CH_2CH_3$), 2.32−2.12 (m, 5H, 2 × $OCOCH_2$ + H-6_{ax}), 2.14 (dd, *J* = 13.0 and 3.5 Hz, H-6_{eq}), 1.61 (m, 4H, 2 × CH_2), 1.53 (m, 2H, CH_2), 0.91 (m, 6H, 2 × CH_3) and 0.80 (t, *J* = 7.5 Hz, 3H, $OCH_2CH_2CH_3$) ppm. ^{13}C NMR (125 MHz, CD_3OD) δ 175.1 (C), 174.7 (C), 174.1 (C), 154.8 (C), 141.3 (C), 140.8 (C), 134.0 (C), 128.3 (CH), 124.9 (CH), 124.9 (CH), 123.6 (CH), 123.5 (CH), 101.9 (CH), 73.5 (C), 71.6 (CH), 71.1 (OCH_2), 70.6 (CH), 68.3 (OCH_2), 37.4 (CH_2), 37.0 (CH_2), 37.0 (CH_2), 23.0 (CH_2), 19.5 (CH_2), 19.4 (CH_2), 13.9 (CH_3), 13.9 (CH_3) and 10.7 (CH_3) ppm. MS (ESI) *m*/*z* (%) 541 (MNa^+). HRMS calcd for $C_{27}H_{34}O_8SNa$ (MNa^+), 541.1867; found, 541.1853.

Propyl (1*R*,4*S*,5*R*)-3-(Benzo[*b*]thiophen-5-ylmethoxy)-4-butyroxy-1,5-dihydroxycyclohex-2-enecarboxylate (8f). The experimental procedure was the same as for compound **8e** using compound **8c** (10 mg), pyridine solution (45 μL), butyryl chloride solution (45 μL), and acetonitrile (0.7 mL). Yield = 3.5 mg (29%). Colorless oil. $[\alpha]_D^{20} = -27°$ (*c* 0.5, MeOH). ^{1}H NMR (400 MHz, CD_3OD) δ 7.87 (d, *J* = 8.4 Hz, 1H, ArH), 7.80 (br s, 1H, ArH), 7.58 (d, *J* = 5.6 Hz, 1H, ArH), 7.35 (d, *J* = 5.6 Hz, 1H, ArH), 7.30 (dd, *J* = 8.4 and 1.6 Hz, 1H, ArH), 5.47 (dd, *J* = 8.0 and 0.8 Hz, 1H, H-4), 5.07 (s, 1H, H-2), 4.90 (s, 2H, OCH_2Ar), 4.19 (m, 1H, H-5), 4.07 (m, 2H, $OCH_2CH_2CH_3$), 2.31 (q, *J* = 7.2 Hz, 2H, $COCH_2$), 2.16 (dd, *J* = 13.2 and 11.2 Hz, 1H, H-6_{ax}), 2.10 (dd, *J* = 13.2 and 4.4 Hz, 1H, H-6_{eq}), 1.62 (m, 2H, CH_2), 1.55 (m, 2H, CH_2), 0.92 (t, *J* = 7.6 Hz, 3H, CH_3) and 0.79 (t, *J* = 7.6 Hz, 3H, $OCH_2CH_2CH_3$) ppm. ^{13}C NMR (100 MHz, CD_3OD) δ 175.9 (C), 175.3 (C), 155.7 (C), 141.3 (C), 140.8 (C), 134.1 (C), 128.2 (CH), 124.9 (CH), 124.9 (CH), 123.5 (CH), 123.4 (CH), 101.1 (CH), 75.0 (CH), 73.8 (C), 71.0 (OCH_2), 68.3 (OCH_2), 68.0 (CH), 40.8 (CH_2), 37.2 (CH_2), 23.0 (CH_2), 19.5 (CH_2), 13.9 (CH_3) and 10.7 (CH_3) ppm. IR (film) 3435 (O−H) and 1651 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 471 (MNa^+). HRMS calcd for $C_{23}H_{28}O_7SNa$ (MNa^+), 471.1448; found, 471.1447.

Esterification of Ethyl Ester 7d: Preparation of Ethyl (1*R*,4*S*,5*R*)-4,5-Dibutyroxy-1-hydroxy-3-(naphth-2-yl)methoxycyclohex-2-ene-1-carboxylate (7f) and Ethyl (1*R*,4*S*,5*R*)-4-Butyroxy-1,5-dihydroxy-3-(naphth-2-yl)methoxycyclohex-2-ene-1-carboxylate (7g). A stirred solution of ethyl ester **7d** (12 mg, 0.06 mmol) in dry acetonitrile (0.8 mL) at 0 °C was treated with a solution of dry pyridine (0.12 mL, 1 M in acetonitrile, 0.12 mmol). After 10 min, a solution of butyryl chloride (0.12 mL, 1 M in acetonitrile, 0.12 mmol) was added. The resulting solution was stirred at room temperature for 10 h and was then partitioned into water and ethyl acetate. The organic layer was separated and the aqueous phase was extracted with ethyl acetate (×3). The combined organic extracts were dried (anhyd $MgSO_4$), filtered, and concentrated under reduced pressure. The residue was purufied by flash chromatography eluting with ethyl acetate/hexane (35:65) to afford ester **7f** (7.3 mg, 42%) and ester **7g** (6.6 mg, 46%), both as colorless oils.

Data for **7f**: $[\alpha]_D^{20} = -29°$ (*c* 1.0, in MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.84 (m, 4H, 4 × ArH), 7.48 (m, 2H, 2 × ArH), 7.44 (dd, *J* = 8.5 and 1.5 Hz, 1H, ArH), 5.65 (dd, *J* = 7.7 and 1.1 Hz, 1H, H-4), 5.46 (m, 1H, H-5), 5.21 (s, 1H, H-2), 4.98 (s, 2H, OCH_2Ar), 4.14 (q, *J* = 7.2 Hz, 2H, OCH_2CH_3), 2.23−2.21 (m, 5H, 2 × CH_2CO + H-6_{ax}), 2.14 (dd, *J* = 13.3 and 4.0 Hz, 1H, H-6_{eq}), 1.65−1.50 (m, 4H, 2 × $COCH_2CH_2CH_3$), 1.21 (t, *J* = 7.2 Hz, 3H, OCH_2CH_3), 0.93 (t, *J* = 7.2 Hz, 3H, $COCH_2CH_2CH_3$) and 0.79 (t, *J* = 7.2 Hz, 3H, $COCH_2CH_2CH_3$) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 175.0 (C), 174.6 (C), 174.1 (C), 154.8 (C), 135.2 (C), 134.7 (C), 134.6 (C), 129.3 (CH), 128.9 (CH), 128.8 (CH), 127.3 (CH), 127.3 (CH), 127.2 (CH), 126.3 (CH), 101.9 (CH), 73.4 (C), 71.6 (CH), 71.0 (OCH_2), 70.6 (CH), 62.8 (OCH_2), 37.4 (CH_2), 37.0 (CH_2), 37.0 (CH_2), 19.5 (CH_2), 19.4 (CH_2), 14.4 (CH_3), 13.9 (CH_3) and 13.9 (CH_3) ppm. IR (film) 3419 (O−H) and 1739 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 521 (MNa^+). HRMS calcd for $C_{28}H_{34}O_8Na$ (MNa^+), 521.2146; found, 521.2136.

Data for **7g**: $[\alpha]_D^{20} = -33°$ (*c* 0.5, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.83 (m, 4H, 4 × ArH), 7.45 (m, 3H, 3 × ArH), 5.49 (d, *J* = 8.2 Hz, 1H, H-4), 5.10 (s, 1H, H-2), 4.94 (s, 2H, OCH_2Ar), 4.25−4.11 (m, 3H, OCH_2CH_3 + H-5), 2.32 (m, 2H, $COCH_2CH_2CH_3$), 2.21−2.11 (m, 2H, CH_2-6), 1.55 (q, *J* = 7.2 Hz, 2H, $COCH_2CH_2CH_3$), 1.21 (t, *J* = 7.0 Hz, 3H, OCH_2CH_3) and 0.78 (t, *J* = 7.2 Hz, 3H, $COCH_2CH_2CH_3$) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 175.7 (C), 175.3 (C), 155.7 (C), 135.3 (C), 134.7 (C), 134.6 (C), 129.2 (CH), 128.9 (CH), 128.7 (CH), 127.3 (CH), 127.2 (CH), 127.1 (CH), 126.3 (CH), 101.1 (CH), 75.0 (CH), 73.7 (C), 70.9 (OCH_2), 68.0 (CH), 62.8 (OCH_2), 40.7 (CH_2), 37.1 (CH_2), 19.5 (CH_2), 14.4 (CH_3) and 13.9 (CH_3) ppm. IR (film) 3446 (O−H) and 1651 (C═O) cm^{-1}. MS (ESI) *m*/*z* (%) 451 (MNa^+). HRMS calcd for $C_{24}H_{28}O_7Na$ (MNa^+), 451.1727; found, 451.1732.

Propyl (1*R*,4*S*,5*R*)-4,5-Dibutyroxy-1-hydroxy-3-(naphth-2-yl)methoxycyclohex-2-ene-1-carboxylate (8g). The experimental procedure was the same as for compound **7f** using compound **8d** (13 mg), 1 M pyridine in acetonitrile (0.12 mL), 1 M butyryl chloride in acetonitrile (0.12 mL), and acetonitrile (0.9 mL). Yield = 11 mg (62%). Colorless oil $[\alpha]_D^{20} = -33°$ (*c* 1.0, MeOH). ^{1}H NMR (250 MHz, CD_3OD) δ 7.83 (m, 4H, 4 × ArH), 7.45 (m, 3H, 3 × ArH), 5.65 (dd, *J* = 7.7 and 0.7 Hz, 1H, H-4), 5.46 (m, 1H, H-5), 5.20 (s, 1H, H-2), 4.98 (s, 2H, OCH_2Ar), 4.04 (t, *J* = 6.5 Hz, 2H, $OCH_2CH_2CH_3$), 2.35−2.24 (m, 5H, 2 × $COCH_2CH_2CH_3$ + H-6_{ax}), 2.14 (dd, *J* = 13.0 and 3.7 Hz, 1H, H-6_{eq}), 1.65−1.50 (m, 6H, 2 × $COCH_2CH_2CH_3$ + $OCH_2CH_2CH_3$), 0.93 (t, *J* = 7.2 Hz, 3H, $OCH_2CH_2CH_3$), 0.88 (t, *J* = 7.5 Hz, 3H, $COCH_2CH_2CH_3$) and 0.80 (t, *J* = 7.2 Hz, 3H, $COCH_2CH_2CH_3$) ppm. ^{13}C NMR (63 MHz, CD_3OD) δ 175.1 (C), 174.6 (C), 174.1 (C), 154.8

(C), 135.2 (C), 134.7 (C), 134.6 (C), 129.3 (CH), 128.9 (CH), 128.8 (CH), 127.3 (CH), 127.3 (CH), 127.2 (CH), 126.2 (CH), 102.0 (CH), 73.4 (C), 71.6 (CH), 71.0 (OCH_2), 70.6 (CH), 68.3 (OCH_2), 37.4 (CH_2), 37.0 (2 × CH_2), 22.9 (CH_2), 19.5 (CH_2), 19.4 (CH_2), 13.9 (CH_3), 13.9 (CH_3) and 10.7 (CH_3) ppm. IR (film) 3433 (O−H) and 1739 (C═O) cm^{-1}. MS (ESI) *m/z* (%) 535 (MNa^+). HRMS calcd for $C_{29}H_{36}O_8Na$ (MNa^+), 535.2302; found, 535.2292.

Dehydroquinase Assays. The enzyme was purified and assayed as described previously.[27,36]

Crystallization of DHQ2/9d and DHQ2/9h Binary Complexes. First, DHQ2-Mt was concentrated to 20 mg mL^{-1} in 50 mM Tris-HCl pH 7.5, 1 mM 2-mercaptoethanol, 1 mM ethylenediaminetetraacetic acid, and 200 mM sodium chloride. A 250 mM solution of inhibitor **9d** or **9h** in methanol was then added at 1:20 (v/v) to give solutions of approximately 10 equiv of inhibitor per protein monomer. Bipyramidal crystals of up to 0.2 mm × 0.2 mm of the DHQ2-Mt/**9d** and **9h** binary complexes were obtained after 3 weeks of vapor diffusion in sitting drops comprised of 2.0 μL of protein/inhibitor solution mixed with 2.0 μL of reservoir solution and equilibrated against 0.15 mL reservoirs containing 33% (v/v) 2-methyl-2,4-pentanediol, 0.3 M ammonium sulfate, and 0.1 M 4-(2-hydroxyethyl)piperazine-1-ethanesulfonic acid sodium salt (HEPES sodium salt), pH 7.5.

Crystallization of DHQ2-Mt/5b Binary Complex. Apo-DHQ2-Mt crystals[12c] were soaked in a 10 mM solution of inhibitor **5b** in the crystallization mixture (32% (v/v) 2-methyl-2,4-pentanediol, 0.3 M ammonium sulfate and 0.1 M 4-(2-hydroxyethyl)-piperazine-1-ethanesulfonic acid sodium salt (HEPES sodium salt), pH 7.5) for 24 h.

Structure Determination of Binary Complexes. Crystals were flash-frozen directly from the crystallization mixtures by rapid immersion in liquid nitrogen. Data were collected on beamline ID23−1, ID23−2, and BM16 (ESRF, Grenoble, France) from crystals maintained at 100 K. The data were processed, scaled, and analyzed using MOSFLM,[37] SCALA,[38] and other programs of the CCP4 software suite.[39] The structure was solved by molecular replacement using the program MOLREP[40] with a search model generated from PDB entry 1H0S.[41] Reflections for calculating R_{free}[42] were selected randomly, and model building and refinement were carried out with COOT[43] and REFMAC,[44] respectively. Structure validation was performed using MOLPROBITY.[45]

The data collection, refinement, and model statistics are summarized in Table 2. Coordinates and structure factors are available from the Protein Data Bank with accession codes 2Y71, 2Y76, and 2Y77 for DHQ2-Mt/**5b**, DHQ2-Mt/**9d**, and DHQ2-Mt/**9h** complexes, respectively. Figures were prepared using PyMOL.[46]

In Vitro Antibacterial Activity Assay. The in vitro antibacterial activity of acids **5** and **9** and their ester derivatives **6**−**11** was studied by determining their minimum inhibitory concentrations (MICs μg/mL) against *M. tuberculosis* H37Rv, using the Alamar Blue Assay.[34] MICs were defined as the lowest concentration at which bacterial growth was no longer evident.

■ ASSOCIATED CONTENT

Supporting Information. Copies of ^1H NMR, ^{13}C NMR, and DEPT spectra for compounds **5**−**11** and Dixon plots for compounds **5** and **9**. This material is available free of charge via the Internet at http://pubs.acs.org.

Accession Codes

‡Coordinates and structure factors are available from the Protein Data Bank with accession codes 2Y71, 2Y76, and 2Y77.

■ AUTHOR INFORMATION

Corresponding Author

*Phone: +34 881815726. Fax: +34 881815704. E-mail: concepcion.gonzalez.bello@usc.es.

■ ACKNOWLEDGMENT

Financial support from the Xunta de Galicia (10PXIB2200-122PR and GRC2010/12) and the Spanish Ministry of Science and Innovation (SAF2010-15076 to C.G.B. and BFU2008-01-588/BMC to M.J.v.R.) is gratefully acknowledged. L.T. and V.F. V.P. thank the Spanish Ministry of Science and Innovation and the Portuguese Funçacão para a Ciência e a Tecnología for FPU and FCT fellowships, respectively. J.M.O. thanks the Xunta de Galicia and the Spanish Ministry of Science and Innovation for "Ángeles Alvariño" and "José Castillejo" fellowships, respectively. We are also thankful to the ESRF for the provison of beam time on beamlines ID23-1, ID23-2 and BM16.

■ DEDICATION

†In memory of Professor Rafael Suau.

■ ABBREVIATIONS USED

TB, tuberculosis; MDR-TB, multidrug-resistant isolates of *Mycobacterium tuberculosis*; XDR-TB, drug-resistant isolates of *Mycobacterium tuberculosis*; DHQ, 3-dehydroquinate dehydratase or dehydroquinase; DHQ1, type I dehydroquinase; DHQ2, type II dehydroquinase; DHQ2-Mt, type II dehydroquinase from *Mycobacterium tuberculosis*; DHQ2-Hp, type II dehydroquinase from *Helicobacter pylori*; PDB, Protein Data Bank; KHMDS, potassium hexamethyldisilazane; LHMDS, lithium hexamethyldisilazane; TBS, *tert*-butyldimethylsilyl; TBAF, tetrabutylammonium fluoride; DMF, *N*,*N*-dimethylformamide; THF, tetrahydrofuran; DTT, 1,4-dithiothreitol; Tris, tris(hydroxymethyl)aminomethane

■ REFERENCES

(1) *Tuberculosis*; World Health Organization: Geneva, 2011; http://www.who.int/tb/research/en/.

(2) (a) Espinal, M. A. The global situation of MDR-TB. *Tuberculosis* **2003**, *83*, 44–51. (b) Dye, C.; Williams, B. G. The population dynamics and control of tuberculosis. *Science* **2010**, *328*, 856–861.

(3) Centers for Disease Control Prevention. Emergence of *Mycobacterium tuberculosis* with extensive resistance to second-line drugs-worldwide, 2000−2004. *MMWR Recomm. Rep.* **2006**, *55*, 301–305.

(4) Raviglione, M. C.; Smith, I. XDR tuberculosis-implications for global public health. *New Engl. J. Med.* **2007**, *356*, 656–659.

(5) Haydel, S. E. Extensively Drug-Resistant Tuberculosis: A Sign of the Times and an Impetus for Antimicrobial Discovery. *Pharmaceuticals* **2010**, *3*, 2268–2290.

(6) Haydel, S. E. Extensively Drug-Resistant Tuberculosis: A Sign of the Times and an Impetus for Antimicrobial Discovery. *Pharmaceuticals* **2010**, *3*, 2268–2290.

(7) Zhang, Y. The magic bullets and tuberculosis drug targets. *Annu. Rev. Pharmacol. Toxicol.* **2005**, *45*, 529–564.

(8) Koul, A.; Arnoult, E.; Lounis, N.; Guillemont, J.; Andries, K. The challenge of new drug discovery for tuberculosis. *Nature* **2011**, *469*, 483–490.

(9) (a) Roberts, F.; Roberts, C. W.; Johnson, J. J.; Kyle, D. E.; Krell, T.; Coggins, J. R.; Coombs, G. H.; Milhous, W. K.; Tzipori, S.; Ferguson, D. J.; Chakrabarti, D.; McLeod, R. Evidence for the shikimate pathway in apicomplexan parasites. *Nature* **1998**, *393*, 801–805. (b) Campbell, S. A.; Richards, T. A.; Mui, E. J.; Samuel, B. U.; Coggins, J. R.; McLeod,

R.; Roberts, C. W. A complete shikimate pathway in *Toxoplasma gondii*: an ancient eukaryotic innovation. *Int. J. Parasit.* **2004**, *34*, 5–13.

(10) Abell, C. Enzymology and Molecular Biology of the Shikimate Pathway. In *Comprehensive Natural Products Chemistry*; Sankawa, U., Ed.; Pergamon, Elsevier Science Ltd.: Oxford, 1999; p 573.

(11) (a) Parish, T.; Stoker, N. G. The common aromatic amino acid biosynthesis pathway is essential in *Mycobacterium tuberculosis*. *Microbiology* **2002**, *148*, 3069–3077.(b) Data base for essential genes in bacteria, see www.essentialgene.org. (c) Zhang, R.; Lin, Y. DEG 5.0, a database of essential genes in both prokaryotes and eukaryotes. *Nucleic Acids Res.* **2009**, *37*, D455–D458. (d) Zhang, R.; Ou, H.-Y.; Zhang, C.-T. DEG, a database of essential genes. *Nucleic Acids Res.* **2004**, *32*, D271–D272. (e) Sassetti, C. M.; Boyd, D. H.; Rubin, E. J. Genes required for mycobacterial growth defined by high density mutagenesis. *Mol. Microbiol.* **2003**, *48*, 77–84.

(12) (a) Shneier, A.; Harris, J.; Kleanthous, C.; Coggins, J. R.; Hawkins, A. R.; Abell, C. Evidence for opposite stereochemical courses for the reactions catalyzed by type I and type II dehydroquinases. *Bioorg. Med. Chem. Lett.* **1993**, *3*, 1399–1402. (b) Harris, J.; Kleanthous, C.; Coggins, J. R.; Hawkins, A. R.; Abell, C. Different mechanistic and stereochemical courses for the reactions catalysed by type I and type II dehydroquinases. *J. Chem. Soc., Chem. Commun.* **1993**, *13*, 1080–1081. (c) Gourley, D. G.; Shrive, A. K.; Polikarpov, I.; Krell, T.; Coggins, J. R.; Hawkins, A. R.; Issacs, N. W.; Sawyer, L. The two types of 3-dehydroquinase have distint structures but catalyze the same overall reaction. *Nature Struct. Biol.* **1999**, *6*, 521–525.

(13) (a) Chauduri, C.; Ducan, K.; Graham, L. D.; Coggins, J. R. Identification of the active-site lysine residues of two biosynthetic 3-dehydroquinases. *Biochem. J.* **1990**, *275*, 1–6. (b) Shneier, A.; Kleanthous, C.; Deka, R.; Coggins, J. R.; Abell, C. Observation of an imine intermediate on dehydroquinase by electrospray mass spectrometry. *J. Am. Chem. Soc.* **1991**, *113*, 9416–9418.

(14) (a) Harris, J.; Kleanthous, C.; Coggins, J. R.; Hawkins, J. R.; Abell, C. Different mechanistic and stereochemical courses for the reactions catalysed by type I and type II dehydroquinases. *J. Chem. Soc., Chem. Commun.* **1993**, 1080–1081. (b) Shneier, A.; Harris, J.; Kleanthous, C.; Coggins, J. R.; Hawkins, A. R.; Abell, C. Evidence for opposite stereochemical courses for the reaction catalysed by type I and type II dehydroquinases. *Bioorg. Med. Chem. Lett.* **1993**, *3*, 1399–1402.

(15) (a) Krell, T.; Pitt, A. R.; Coggins, J. R. The use of electrospray mass spectrometry to identify an essential arginine residue in type II dehydroquinases. *FEBS Lett.* **1995**, *360*, 93–96. (b) Krell, T.; Horsburgh, M. J.; Cooper, A.; Kelly, S. M.; Coggins, J. R. Localization of the active site of type II dehydroquinase. Identification of a common arginine-containing motif in the two classes of dehydroquinases. *J. Biol. Chem.* **1996**, *271*, 24492–24497.

(16) Roszak, A. W.; Robinson, D. A.; Krell, T.; Hunter, I. S.; Frederickson, M.; Abell, C.; Coggins, J. R.; Lapthorn, A. J. The structure and mechanism of the type II dehydroquinase from *Streptomyces coelicolor*. *Structure* **2002**, *10*, 493–503.

(17) Peón, A.; Otero, J. M.; Tizón, L.; Prazeres, V. F. V.; Llamas-Saiz, A. L.; Fox, G. C.; van Raaij, M. J.; Lamb, H.; Hawkins, A. R.; Gago, F.; Castedo, L.; González-Bello, C. Understanding the key factors that control the inhibition of type II dehydroquinase by (2*R*)-2-benzyl-3-dehydroquinic acids. *ChemMedChem* **2010**, *5*, 1726–1733.

(18) Gonzalez-Bello, C.; Castedo, L. Progress in type II dehydroquinase inhibitors: from concept to practice. *Med. Res. Rev.* **2007**, *27*, 177–208.

(19) Frederickson, M.; Parker, E. J.; Hawkins, A. R.; Coggins, J. R.; Abell, C. Selective inhibition of type II dehydroquinases. *J. Org. Chem.* **1999**, *64*, 2612–2613.

(20) González-Bello, C.; Lence, E.; Toscano, M. D.; Castedo, L.; Coggins, J. R.; Abell, C. Parallel solid-phase synthesis and evaluation of inhibitors of *Streptomyces coelicolor* type II dehydroquinase. *J. Med. Chem.* **2003**, *46*, 5735–5744.

(21) Frederickson, M.; Roszak, A. W.; Coggins, J. R.; Lapthorn, A. J.; Abell, C. (1*R*,4*S*,5*R*)-3-Fluoro-1,4,5-trihydroxy-2-cyclohexene-1-carboxylic acid: the fluoro analogue of the enolate intermediate in the reaction catalyzed by type II dehydroquinases. *Org. Biomol. Chem.* **2004**, *2*, 1592–1596.

(22) Sánchez-Sixto, C.; Prazeres, V. F. V.; Castedo, L.; Lamb, H.; Hawkins, A. R.; González-Bello, C. Structure-based design, synthesis and biological evaluation of inhibitors of *Mycobacterium tuberculosis* type II dehydroquinase. *J. Med. Chem.* **2005**, *48*, 4871–4881.

(23) Prazeres, V. F. V.; Sánchez-Sixto, C.; Castedo, L.; Lamb, H.; Hawkins, A. R.; Riboldi-Tunnicliffe, A.; Coggins, J. R.; Lapthorn, A. J.; González-Bello, C. Nanomolar mechanism-based inhibitors of *Mycobacterium tuberculosis* and *Streptomyces coelicolor* type II dehydroquinase. *ChemMedChem* **2007**, *2*, 194–207.

(24) Payne, R. J.; Riboldi-Tunnicliffe, A. L.; Kerbarh, V. V. V.; Abell, A. D.; Lapthorn, A. J.; Abell, C. Design, synthesis, and structural studies on potent biaryl inhibitors of type II dehydroquinases. *ChemMedChem* **2007**, *2*, 1010–1013.

(25) Toscano, M. D.; Payne, R. J.; Chiba, A.; Kerbarh, O.; Abell, C. Nanomolar inhibition of type II dehydroquinase based on the enolate reaction mechanism. *ChemMedChem* **2007**, *2*, 101–112.

(26) Payne, R. J.; Peyrot, F.; Kerbarh, O.; Abell, A. D.; Abell, C. Rational design, synthesis, and evaluation of nanomolar type II dehydroquinase inhibitors. *ChemMedChem* **2007**, *2*, 1015–1029.

(27) Prazeres, V. F. V.; Sánchez-Sixto, C.; Castedo, L.; Shuh, S. W.; Lamb, H.; Hawkins, A. R.; Cañada, F. J.; Jiménez-Barbero, J.; González-Bello, C. Competitive inhibitors of *Helicobacter pylori* type II dehydroquinase: synthesis, biological evaluation, and NMR studies. *ChemMedChem* **2008**, *3*, 756–770.

(28) Prazeres, V. F. V.; Tizón, L.; Otero, J. M.; Guardado-Calvo, P.; Llamas-Saíz, A. L.; van Raaij, M. J.; Castedo, L.; Lamb, H.; Hawkins, A. R.; González-Bello, C. Synthesis and biological evaluation of new nanomolar competitive inhibitors of *Helicobacter pylori* type II dehydroquinase. Structural details of the role of the aromatic moieties with essential residues. *J. Med. Chem.* **2010**, *53*, 191–200.

(29) Paz, S.; Tizón, L.; Otero, J. M.; Llamas-Saiz, A. L.; Fox, G. C.; van Raaij, H. J.; Lamb, H.; Hawkins, A. R.; Lapthorn, A. J.; Castedo, L.; González-Bello, C. Tetrahydrobenzothiophene derivatives: conformationally restricted inhibitors of type II dehydroquinase. *ChemMedChem* **2011**, *6*, 266–272.

(30) Tran, A. T.; Cergol, K. M.; Britton, W. J.; Bokhari, S. A. I.; Ibraim, M.; Lapthorn, A. J.; Payne, R. J. Rapid assembly of potent type II dehydroquinase inhibitors via click chemistry. *Med. Chem. Commun.* **2010**, *1*, 271–275.

(31) Prazeres, V. F. V.; Sánchez-Sixto, C.; Castedo, L.; Canales, A.; Cañada, F. J.; Jiménez-Barbero, J.; Lamb, H.; Hawkins, A. R.; González-Bello, C. Determination of the bound conformation of a competitive nanomolar inhibitor of *Mycobacterium tuberculosis* type II dehydroquinase by NMR spectroscopy. *ChemMedChem* **2006**, *1*, 990–996.

(32) Prazeres, V. F. V.; Castedo, L.; Lamb, H.; Hawkins, A. R.; González-Bello, C. 2-Substituted-3-dehydroquinic acids as potent competitive inhibitors of type II dehydroquinase. *ChemMedChem* **2009**, *4*, 1980–1984.

(33) The X-ray crystal structure is available from the Protein Data Bank (PBD: 1H0S): Robinson, D. A.; Roszak, A. W.; Frederickson, M.; Abell, C.; Coggins, J. R.; Lapthorn, A. J. Structural basis for specificity of oxime based inhibitors towards type II dehydroquinase from *M. tuberculosis*. (to be published) Residues 20−25 are not visible, including the essential residue Tyr24.

(34) Palomino, J. C.; Martin, A.; Camacho, M.; Guerra, H.; Swings, J.; Portaels, F. Resazurin microtiter assay plate: simple and inexpensive method for detection of drug resistance in *Mycobacterium tuberculosis*. *Antimicrob. Agents Chemother.* **2002**, *46*, 2720–2722.

(35) Charman, W. N.; Porter, C. J. H. Lipophilic prodrugs designed for intestinal lymphatic transport. *Adv. Drug Delivery Rev.* **1996**, *19*, 149–169.

(36) Gourley, D. G.; Coggins, J. R.; Isaacs, N. W.; Moore, J. D.; Charles, I. G.; Hawkins, A. R. Crystallization of a type II dehydroquinase from *Mycobacterium tuberculosis*. *J. Mol. Biol.* **1994**, *241*, 488–491.

(37) Leslie, A. G. The integration of macromolecular diffraction data. *Acta Crystallogr., Sect. D: Biol. Crystallogr.* **2006**, *62*, 48–57.

(38) Evans, P. Scaling and assessment of data quality. *Acta Crystallogr., Sect. D: Biol. Crystallogr.* **2006**, *62*, 72–82.
(39) Winn, M. D. An overview of the CCP4 project in protein crystallography: an example of a collaborative project. *J. Synchrotron Radiat.* **2003**, *10*, 23–25.
(40) Vagin, A.; Teplyakov, A. MOLREP: an automated program for molecular replacement. *J. Appl. Crystallogr.* **1997**, *30*, 1022–1025.
(41) The X-ray crystal structure is available from the Protein Data Bank (PBD: 1H0S): Robinson, D. A.; Roszak, A. W.; Frederickson, M.; Abell, C.; Coggins, J. R.; Lapthorn, A. J. Structural basis for specificity of oxime based inhibitors towards type II dehydroquinase from *M. tuberculosis*. (to be published) Residues 20−25 are not visible, including the essential residue Tyr24.
(42) Brunger, A. T. Free *R* value: cross-validation in crystallography. *Methods Enzymol.* **1997**, *277*, 366–396.
(43) Emsley, P.; Cowtan, K. Coot: model-building tools for molecular graphics. *Acta Crystallogr., Sect. D: Biol. Crystallogr.* **2004**, *60*, 2126–2132.
(44) Murshudov, G. N.; Vagin, A. A.; Dodson, E. J. Refinement of macromolecular structures by the maximum-likelihood method. *Acta Crystallogr., Sect. D: Biol. Crystallogr.* **1997**, *53*, 240–255.
(45) Chen, V. B.; Arendall, W. B., III; Headd, J. J.; Keedy, D. A.; Immormino, R. M.; Kapral, G. J.; Murray, L. W.; Richardson, J. S.; Richardson, D. C. MolProbity: all-atom structure validation for macromolecular crystallography. *Acta Crystallogr., Sect. D: Biol. Crystallogr.* **2010**, *D66*, 12–21.
(46) DeLano, W. L. *The PyMOL Molecular Graphics System*; DeLano Scientific LLC: Palo Alto, CA, 2008; http://www.pymol.org.
(47) Davis, I. W.; Leaver-Fay, A.; Chen, V. B.; Block, J. N.; Kapral, G. J.; Wang, X.; Murray, L. W.; Arendall, W. B., 3rd; Snoeyink, J.; Richardson, J. S.; Ricchardson, D. C. MolProbity: all-atom contacts and structure validation for proteins and nucleic acids. *Nucleic Acids Res.* **2007**, *35*, W375–W383.