J|A|C|S ARTICLES

Published on Web 03/30/2009

Structure Proof and Synthesis of Kotalanol and De-*O*-sulfonated Kotalanol, Glycosidase Inhibitors Isolated from an Herbal Remedy for the Treatment of Type-2 Diabetes

Kumarasamy Jayakanthan, Sankar Mohan, and B. Mario Pinto*

Department of Chemistry, Simon Fraser University, Burnaby, British Columbia, Canada V5A 1S6

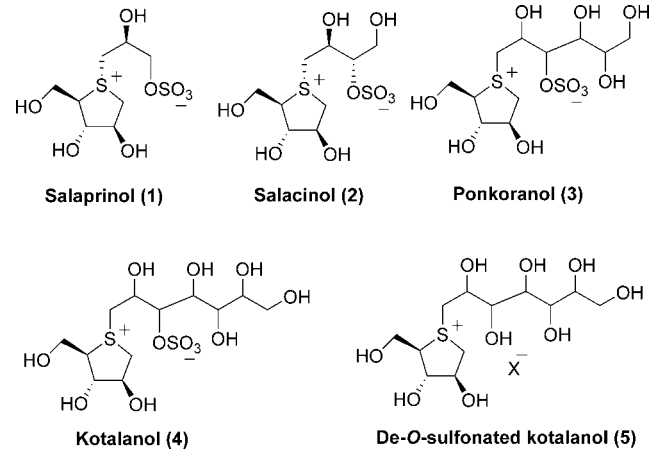
Received February 4, 2009; E-mail: bpinto@sfu.ca

Abstract: Kotalanol and de-*O*-sulfonated-kotalanol are the most active principles in the aqueous extracts of *Salacia reticulata* which are traditionally used in India, Sri Lanka, and Thailand for the treatment of diabetes. We report here the exact stereochemical structures of these two compounds by synthesis and comparison of their physical data to those of the corresponding natural compounds. The candidate structures were based on our recent report on the synthesis of analogues and also the structure−activity relationship studies of lower homologues. The initial synthetic strategy relied on the selective nucleophilic attack of *p*-methoxybenzyl (PMB)-protected 4-thio-D-arabinitol at the least hindered carbon atom of two different, selectively protected 1,3-cyclic sulfates to afford the sulfonium sulfates. The protecting groups consisted of a methylene acetal, in the form of a seven-membered ring, and benzyl ethers. Deprotection of the adducts yielded the sulfonium ions but also resulted in de-*O*-sulfonation. Comparison of the physical data of the two adducts to those reported for de-*O*-sulfonated natural kotalanol yielded the elusive structure of kotalanol by inference. The side chain of this compound was determined to be another naturally occurring heptitol, D-perseitol (D-glycero-D-galacto-heptitol) with a sulfonyloxy group at the C-5 position. The synthesis of kotalanol itself was then achieved by coupling PMB-protected 4-thio-D-arabinitol with a cyclic sulfate that was synthesized from the naturally occurring D-perseitol. The work establishes unambiguously the structures of two natural products, namely, kotalanol and de-*O*-sulfonated kotalanol.

Introduction

Glycosidase inhibitors can be used to probe biological recognition processes in cell−cell or cell−virus interactions, as components of diagnostic agents, or as therapeutic agents.[1] The isolation of new candidates from natural sources coupled with their synthesis has attracted a great deal of attention.[2] Of particular relevance to the present work, Yoshikawa et al. discovered a new class of glycosidase inhibitors, namely, salaprinol (**1**),[3] salacinol (**2**),[4] ponkoranol (**3**),[3] and kotalanol (**4**)[5] from the plant *Salacia reticulata*, all of which possess a common sulfonium ion stabilized with an internal sulfate counterion and differing only in the number of carbons in the polyhydroxylated side chain (Chart 1). Recently, Ozaki et al.[6] isolated another α-glucosidase inhibitor from the same plant and assigned its structure to a 13-membered cyclic sulfoxide; this structure has been reassigned by Yoshikawa et al.[7] to be the de-*O*-sulfonated kotalanol (**5**). The latter compound was shown by Ozaki et al.[6] to be the most active compound against

Chart 1



(1) (a) Naturally occurring glycosidase inhibitors. In *Bioactive Natural Products*, 2nd ed.; Nash, R. J., Ed.; Summit Wales Limited: Plas Gogerddan, Aberystwyth, U.K., 2008; pp 407−420. (b) Rempel, B. P.; Withers, S. G. *Glycobiology* **2008**, *18*, 570–586. (c) Borges de Melo, E.; Gomes, A.; Carvalho, I. *Tetrahedron* **2006**, *62*, 10277–10302. (d) Lopez, O.; Bols, M. *Iminosugars* **2007**, 131–151. (e) Asano, N. *Glycobiology* **2003**, *13*, 93R–104R.

(2) (a) Watson, A. A.; Fleet, G. W. J.; Asano, N.; Molyneux, R. J.; Nash, R. J. *Phytochemistry* **2001**, *56*, 265–295. (b) Siriwardena, A.; Strachan, H.; El-Dahar, S.; Way, G.; Winchester, B.; Glushka, J.; Moreman, K.; Boons, G.-J. *ChemBioChem* **2005**, *6*, 845–848. (c) Asano, N.; Nash, R. J.; Molyneux, R. J.; Fleet, G. W. J. *Tetrahedron: Asymmetry* **2000**, *11*, 1645–1680. (d) Nishimura, Y. *Heterocycles* **2006**, *67*, 461–488. (e) Tatsuta, K. *Curr. Org. Chem.* **2001**, *5*, 207–231.

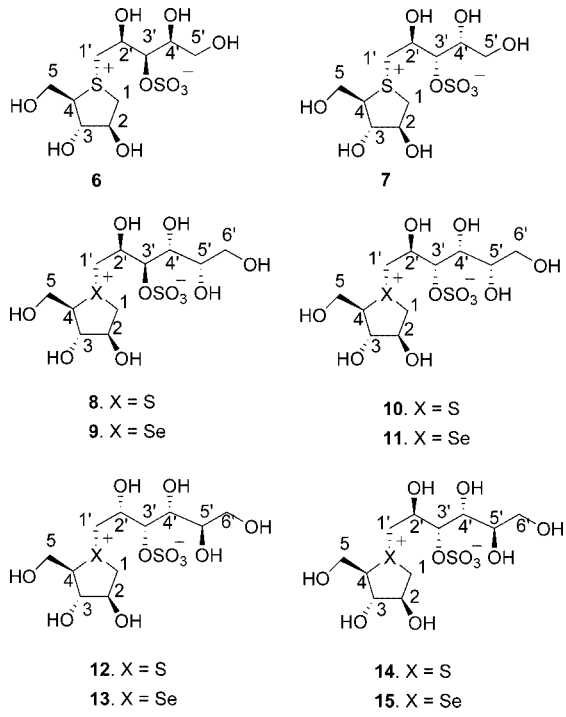
(3) Yoshikawa, M.; Xu, F.; Nakamura, S.; Wang, T.; Matsuda, H.; Tanabe, G.; Muraoka, O. *Heterocycles* **2008**, *75*, 1397–1405.

(4) Yoshikawa, M.; Murakami, T.; Shimada, H.; Matsuda, H.; Yomahara, J.; Tanabe, G.; Muraoka, O. *Tetrahedron Lett.* **1997**, *38*, 8367–8370.

(5) Yoshikawa, M.; Murakami, T.; Yashiro, K.; Matsuda, H. *Chem. Pharm. Bull.* **1998**, *46*, 1339–1340.

(6) (a) Ozaki, S.; Oe, H.; Kitamura, S. *J. Nat. Prod* **2008**, *71*, 981–984. (b) Oe, H.; Ozaki, S. *Biosci. Biotechnol. Biochem.* **2008**, *72*, 1962–1964.

Chart 2



rat intestinal glucosidase in this series of compounds (compare the K_i values for salacinol (0.97, 0.20, and 1.1 μM), kotalanol (0.54, 0.42, and 4.2 μM), and de-*O*-sulfonated kotalanol (0.11, 0.05, and 0.42 μM) using maltose, sucrose, and isomaltose as substrates, respectively). It is noteworthy that aqueous extracts of the roots and stems of the plant *S. reticulata* have been traditionally used in the Ayurvedic system of Indian medicine for the treatment of type-2 diabetes. Recent clinical trials on human patients with type-2 diabetes mellitus using the aqueous extract of the same plant have indicated good glycemic control and side effects comparable to the placebo control group.[8] We and others have carried out extensive research on the synthesis of salacinol (**2**) and higher homologues, differing in stereochemistry at the stereogenic centers, and congeners in which the sulfur heteroatom has been substituted by the cognate atoms nitrogen and selenium.[9] Interestingly, one of our synthetic compounds[10] (**10**, Chart 2), a six-carbon-chain homologue of salacinol, was recently isolated from the same plant and assigned the name ponkoranol (**3**).[3] Since the exact stereochemical structure of kotalanol (**4**) is not yet known, we have now turned our attention to its determination.

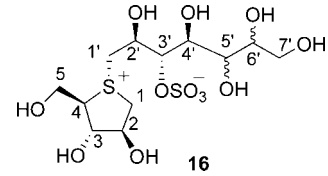
Chemical degradation studies indicated that the 1-deoxy-4-thiopentofuranosyl portion of kotalanol is identical to that in salacinol.[5] However, the absolute configuration of the stereogenic centers in the heptitol side chain and at the sulfur center has not yet been determined. Our previous synthetic work has yielded several five-carbon- and six-carbon-chain analogues as well as selenium congeners **6**−**15** (Chart 2) which have been screened for inhibitory activity against recombinant human

Table 1. Experimentally Determined K_i Values[a]

	stereochemistry of the acyclic side chain				
inhibitor	C-2′	C-3′	C-4′	C-5′	K_i (μM)
6	*S*	*R*	*S*		NA[b10]
7	S	*S*	*R*		0.26 ± 0.02[10]
8	*S*	*R*	*R*	*S*	0.25 ± 0.02[10]
9	*S*	*R*	*R*	*S*	0.10 ± 0.02[11]
10	*S*	*S*	*R*	*S*	0.17 ± 0.03[10]
11	*S*	*S*	*R*	*S*	0.10 ± 0.02[11]
12	*R*	*S*	*R*	*R*	NA[b12]
13	*R*	*S*	*R*	*R*	41.0 ± 7.0[12]
14	*S*	*S*	*R*	*R*	0.65 ± 0.10[13]
15	*S*	*S*	*R*	*R*	0.14 ± 0.03[13]
salacinol	*S*	*S*			0.19 ± 0.02[14]
blintol	*S*	*S*			0.49 ± 0.05[14]

[a] Analysis of MGA inhibition was performed using maltose as the substrate and measuring the release of glucose. Absorbance measurements were averaged to give a final result. [b] NA: not active.

Chart 3

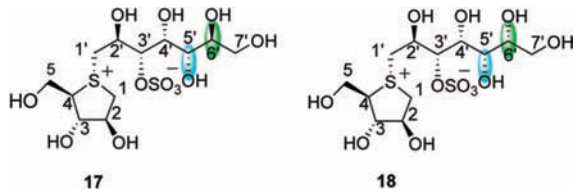


maltase glucoamylase (MGA), a critical intestinal glucosidase involved in the breakdown of glucose oligomers into glucose. Several of the compounds showed inhibitory activity in the low micromolar range (Table 1). It is clear that the stereochemistry at the different stereogenic centers on the side chain plays a significant role in biological activity. It appears that the compounds containing the *S* configuration at C-2′, the *R* configuration at C-4′, and the *S* configuration at C-5′ are the most active in the sulfur series of compounds; we note, however, that in the selenium series the activities of the selenium analogues, **11** and **15** (0.10 and 0.14), suggest that the stereochemistry at C-5′ could be *R*. The stereochemistry at C-3′ was judged to be unimportant but can be fixed as *S* to reflect a presumed common biosynthetic pathway as salacinol. It is noteworthy that each of the seven carbon analogues (Chart 3, **16**) recently synthesized by Muraoka et al.[15] with the *S*-configuration at C-4′ showed less inhibitory activity than natural kotalanol (**4**), also suggesting that the necessary stereochemistry at C-4′ is *R*.

On the basis of these data, we previously proposed the structures **17** or **18** for kotalanol (Chart 4).[16] These candidates were synthesized, and although they showed inhibitory values

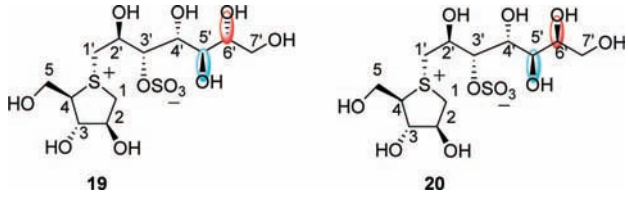
(7) Muraoka, O.; Xie, W.; Tanabe, G.; Amer, M. F. A.; Minematsu, T.; Yoshikawa, M. *Tetrahedron Lett.* **2008**, *49*, 7315–7317.
(8) Jayawardena, M. H. S.; de Alwis, N. M. W.; Hettigoda, V.; Fernando, D. J. S. *J. Ethnopharmacol.* **2005**, *97*, 215–218.
(9) For a review, see: Mohan, S.; Pinto, B. M. *Carbohydr. Res.* **2007**, *342*, 1551–1580.
(10) Johnston, B. D.; Jensen, H. H.; Pinto, B. M. *J. Org. Chem.* **2006**, *71*, 1111–1118.
(11) Liu, H.; Nasi, R.; Jayakanthan, K.; Sim, L.; Heipel, H.; Rose, D. R.; Pinto, B. M. *J. Org. Chem.* **2007**, *72*, 6562–6572.
(12) (a) Liu, H.; Pinto, B. M. *Can. J. Chem.* **2006**, *84*, 1351–1362. (b) Liu, H.; Sim, L.; Rose, D. R.; Pinto, B. M. *J. Org. Chem.* **2006**, *71*, 3007–3013.
(13) Nasi, R.; Sim, L.; Rose, D. R.; Pinto, B. M. *J. Org. Chem.* **2007**, *72*, 180–186.
(14) Rossi, E. J.; Sim, L.; Kuntz, D. A.; Hahn, D.; Johnston, B. D.; Ghavami, A.; Szczepina, M. G.; Kumar, N. S.; Strerchi, E. E.; Nichols, B. L.; Pinto, B. M.; Rose, D. R. *FEBS J.* **2006**, *273*, 2673–2683.
(15) (a) Tanabe, G.; Matsuoka, K.; Minematsu, T.; Morikawa, T.; Ninomiya, K.; Matsuda, H.; Yoshikawa, M.; Murata, H.; Muraoka, O. *J. Pharm. Soc. Jpn.* **2007**, *127* (Suppl. 4), 129–130. (b) Muraoka, O.; Tanabe, G. World Intellectual Property Organization Patent, Pub. No. WO/2008/082017, International Application No. PCT/JP2008/050223, 2008.
(16) Nasi, R.; Patrick, B. O.; Sim, L.; Rose, D. R.; Pinto, B. M. *J. Org. Chem.* **2008**, *73*, 6172–6181.

Chart 4



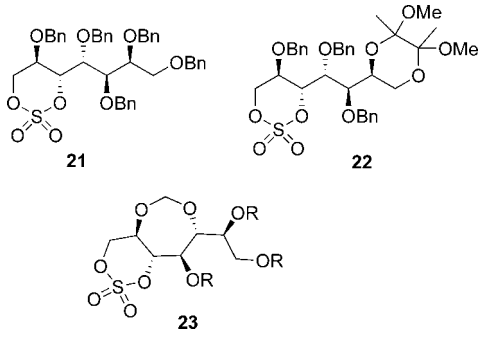
17 **18**

Chart 5



19 **20**

Chart 6



21 **22** **23**

in the low micromolar range, comparable to kotalanol, the discrepancies in the ^{1}H and ^{13}C NMR data indicated that **17** or **18** was not in fact kotalanol.

Detailed inspection of the NMR data of **17**, **18**, and kotalanol also led us to predict that the configuration at C-5′ in the natural product is *R*.[16] We therefore embarked on the synthesis of compounds **19** and **20**, with the *S* configuration at C-2′ and C-3′, the *R* configuration at C-4′ and C-5′, and either the *S* or *R* configuration at C-6′ in order to determine the exact stereochemistry of kotalanol.

Results and Discussion

These two analogues **19** and **20** (Chart 5) could be obtained by alkylation of a thioether with terminal 1,3-cyclic sulfates (Chart 6). The heptitol-derived cyclic sulfates could be synthesized, in turn, from a hexose by successive Wittig and asymmetric dihydroxylation reactions. The desired stereochemistry at C-2′−C-5′ could be readily introduced from D-mannitol.

Our initial attempts employed the cyclic sulfates **21** and **22** (Chart 6), but intramolecular ring opening of the cyclic sulfate moiety by one of the benzyl ethers caused decomposition. Therefore, we decided to introduce some rigidity to the cyclic sulfate through protecting groups in order to avoid the intramolecular ring-opening reaction. Our previous work also suggested that release of torsional strain in the cyclic sulfate led to increased reactivity.[12] Accordingly, we chose the methylene acetal (see **23**) as a protecting group which could survive the acidic conditions required for removal of the benzylidene acetal prior to installation of the cyclic sulfate; the methylene acetal can be introduced under strongly basic conditions.

Thus, di-*O*-benzylidene-D-mannitol, **24**,[17] was treated with dibromomethane in the presence of aqueous sodium hydroxide and tetra-*n*-butylammonium bromide as catalyst;[18] removal of one of the benzylidene groups using catalytic *p*-toluenesulfonic acid (PTSA) in methanol then gave the diol **25**[19] in 65% yield over two steps (Scheme 1). In the deprotection reaction, owing to the C-2-symmetric nature of compound **24**, removal of either benzylidene group led to the same diol, **25**. The primary hydroxyl group was selectively protected with *tert*-butyldimethylsilyl chloride (TBDMS) followed by protection of the secondary hydroxyl group as its benzyl ether. Finally, the silyl protecting group was removed using tetra-*n*-butylammonium fluoride to yield **26** in 62% yield over three steps. Oxidation of the alcohol **26** using Dess−Martin periodinane gave the aldehyde, which was treated with methyltriphenylphosphonium bromide to yield the olefin **27** in 56% yield over two steps.

Kishi's empirical rule for dihydroxylation of acyclic allylic alcohols[20] suggests that treatment of the olefin **27** with OsO_4 should yield the *syn*-dihydroxylated product, with the *erythro* configuration between the pre-existing hydroxyl group and the newly generated hydroxyl group. Sharpless asymmetric dihydroxylation[21] using AD-mix-β should offer the other diastereomer. Thus, treatment of the olefin **27** under OsO_4-catalyzed dihydroxylation conditions gave a diastereomeric ratio of 7:1, with the major isomer **28** in 84% yield (Scheme 2). The major isomer was separated by column chromatography, and then the hydroxyl groups were protected as benzyl ethers. To introduce the cyclic sulfate moiety, the benzylidene group was first removed using catalytic PTSA in methanol, and the resulting diol **29** was then treated with thionyl chloride and triethylamine, followed by oxidation of the corresponding cyclic sulfite with sodium periodate and ruthenium(III) chloride as a catalyst to give the cyclic sulfate **30** in 61% yield.

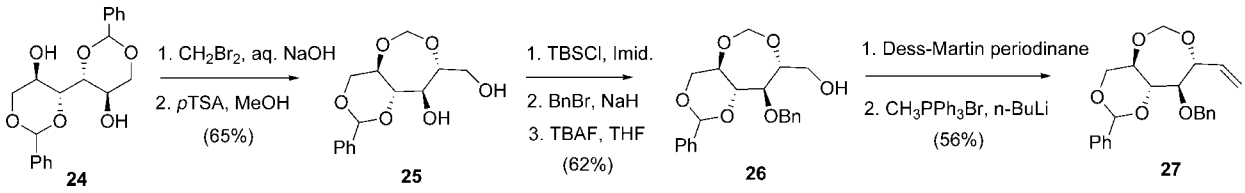
In order to prove the stereochemistry at the newly formed stereogenic center (C-6) of compound **28**, it was converted into the tricyclic derivative **31** as shown in Scheme 3. The observed coupling constant ($J_{5,6}$ = 10.2 Hz) between protons H-5 and H-6 confirms their diaxial relationship and thus proves the configuration at C-6 as being *R*. The assignment was corroborated by the observed NOE contact between H-4 and H-6 (Scheme 3).

In order to obtain the other diastereomer, with the *S* configuration (at C-2), the olefin **27** was treated with AD-mix-β in *tert*-BuOH-H_2O (1:1). A diastereomeric ratio of 7:1 was obtained, with the major isomer **32** in 64% yield. The diol **32** was converted into the corresponding cyclic sulfate **34**, as for the case of **30** (Scheme 4).

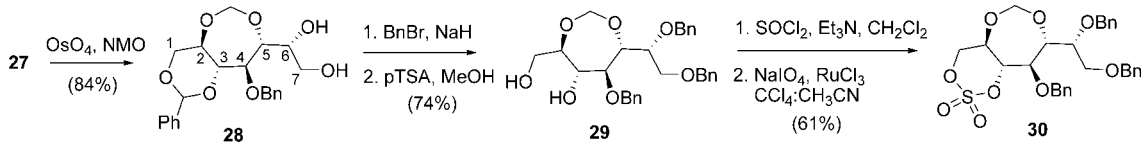
With the cyclic sulfates **30** and **34** in hand, we turned our attention to the coupling reactions with the thio-arabinitol **35**[22] in 1,1,1,3,3,3-haxafluoro-2-propanol (HFIP) as a solvent containing K_2CO_3. The coupling reaction was found to proceed slowly at 75 °C, with some decomposition occurring above 80 °C, so the reactions were terminated after stirring at 75 °C for 7 days to give the corresponding coupled

(17) Baggett, N.; Stribblehill, P. *J. Chem. Soc., Perkin Trans. 1* **1977**, 1123–1126.
(18) Kim, K. S.; Szarek, W. A. *Synthesis* **1978**, 48–50.
(19) Grindley, T. B.; Szarek, W. A. *Can. J. Chem.* **1974**, *52*, 4062–4070.
(20) Harris, J. M.; Keranen, M. D.; O'Doherty, G. A. *J. Org. Chem.* **1999**, *64*, 2982–2983.
(21) Jacobsen, E. N.; Markó, I.; Mungall, W. S.; Schröder, G.; Sharpless, K. B. *J. Am. Chem. Soc.* **1988**, *110*, 1968–1970.
(22) Ghavami, A.; Sadalapur, K. S.; Johnston, B. D.; Lobera, M.; Snider, B. B.; Pinto, B. M. *Synlett* **2003**, 1259–1262.

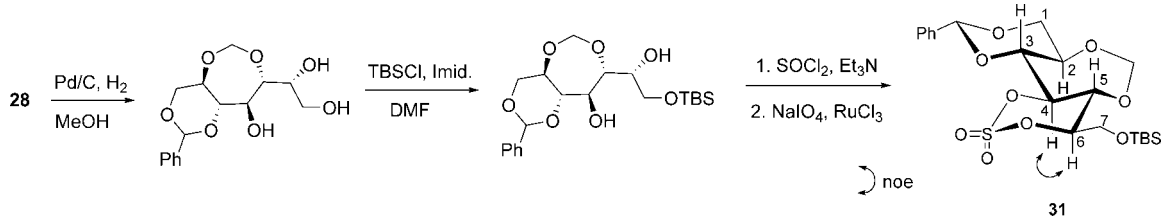
Scheme 1



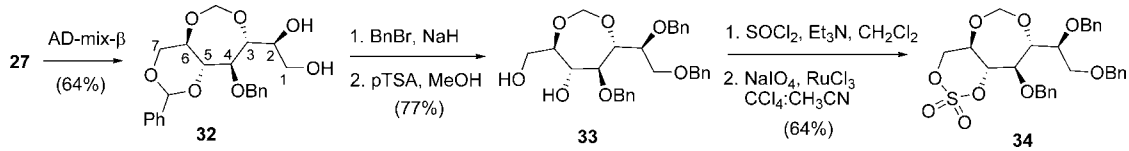
Scheme 2



Scheme 3



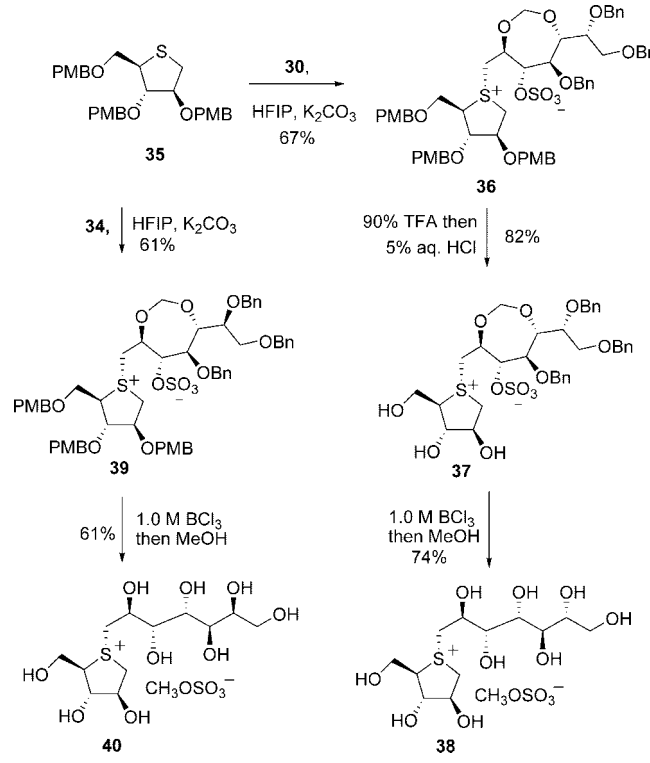
Scheme 4



products **36** and **39** in 67% and 61% yield, respectively (Scheme 5). Finally, deprotection of **36** was carried out by treatment with 90% trifluoroacetic acid in water. Unfortunately, the methylene groups were found to survive these reaction conditions, as well as treatment with 5% aqueous hydrochloric acid at 40 °C, and yielded compound **37**.

Treatment of **37** with 1.0 M BCl_3 in methylene chloride was successful in removing all protecting groups but unfortunately also resulted in desulfonation, thus leading to compound **38**. Similarly, the protected compound **39** was also treated with 1.0 M BCl_3 to yield compound **40**. Interestingly, de-*O*-sulfonated kotalanol **5** has been obtained from kotalanol by Yoshikawa et al.[3] and also isolated recently by Ozaki et al.,[6] as claimed by Yoshikawa et al.[7] With the de-*O*-sulfonated compounds **38** and **40** in hand, it was therefore possible to compare their physical data to those of authentic de-*O*-sulfonated kotalanol. Comparison of the ^1H and ^{13}C NMR data with those of the naturally derived de-*O*-sulfonated kotalanol **5** is shown in Table 2 (for bar graph comparison, see Supporting Information). Our synthetic compounds **38** and **40** have $CH_3OSO_3^-$ as the external counterion, as confirmed by ^1H and ^{13}C NMR spectroscopy. In addition, Yoshikawa et al. reported that the counterion has no significant effect on the NMR chemical shifts.[7] The ^1H and ^{13}C NMR spectra of **38** and **40** were recorded in CD_3OD and assigned unambiguously with the aid of $^1H-^1H$ COSY, HMQC, HMBC,

Scheme 5



and APT experiments. The stereochemistry at the stereogenic sulfonium center in **38** and **40** was established by means of

(23) (a) Montgomery, E. M.; Hudson, C. S. *J. Am. Chem. Soc.* **1939**, *61*, 1654–1658. (b) Nordal, A.; Benson, A. A. *J. Am. Chem. Soc.* **1954**, *76*, 5054–5055. (c) Jones, J. K. N.; Wall, R. A. *Nature* **1961**, *189*, 746–746.

Table 2. Comparison of ^{1}H and ^{13}C NMR Data to Those Reported for De-*O*-sulfonated Kotalanol in CD_3OD

	^{1}H NMR data			^{13}C NMR data		
	38	**5**[3]	**40**	**38**	**5**[3]	**40**
1′	3.94, 3.75	3.94, 3.76	3.94, 3.76	52.7	52.7	52.7
2′	4.17	4.18	4.18	69.4	69.7	69.7
3′	3.85	3.84	3.85	74.0	70.2	70.2
4′	3.88	3.65	3.65	71.9	71.3	71.2
5′	3.71	3.85	3.84	73.1	73.6	73.6
6′	3.83	3.93	3.93	74.8	71.7	71.7
7′	3.80, 3.67	3.66	3.66	64.4	65.0	64.9
1	3.86	3.87	3.87	51.9	51.9	51.9
2	4.62	4.62	4.62	79.4	79.4	79.4
3	4.37	4.37	4.37	79.5	79.5	79.5
4	4.02	4.02	4.01	73.7	73.7	73.7
5	4.05, 3.93	4.05, 3.93	4.05, 3.93	61.1	61.1	61.1

Table 3. Comparison of the Observed Coupling Constants for H-2 and H-6 in the ^{1}H NMR Spectra of Compounds **41**−**44**

	coupling constant[d] (Hz)	
compound	H-2 (J_{1a-2}, J_{1b-2}, J_{2-3})	H-6 (J_{7a-6}, J_{7b-6}, J_{5-6})
41[a]	1.2, 1.2, 1.2	5.4, 10.2, 9.6
42[b]	4.2, 4.2, 1.8	4.8, 10.8, 9.6
43[c]	1.2, 1.2, 1.2	4.2, 4.2, 8.4
44[c]	1.2, 1.2, 1.8	7.2, 7.2, 9.6

[a] Solvent: pyridine-d_5 + CD_3OD. [b] Solvent: $CDCl_3$ + D_2O. [c] Solvent: $CDCl_3$. [d] 600 MHz NMR.

NOESY experiments. A correlation between H-1′ and H-4 confirmed the trans relationship between the side chain and the C-4 substituent of the thio-arabinitol moiety, as found in our previous studies.[9] It is clear from these data that de-*O*-sulfonated kotalanol possesses the structure displayed by **40**. This conclusion is corroborated by the optical rotation data [**38** $[\alpha]_D^{23}$ −4.0 (c = 0.8, MeOH); **40** $[\alpha]_D^{23}$ +10.0 (c = 0.6, MeOH); naturally derived desulfonated kotalanol **5** $[\alpha]_D^{23}$ +13.0 (c = 0.6, MeOH)].

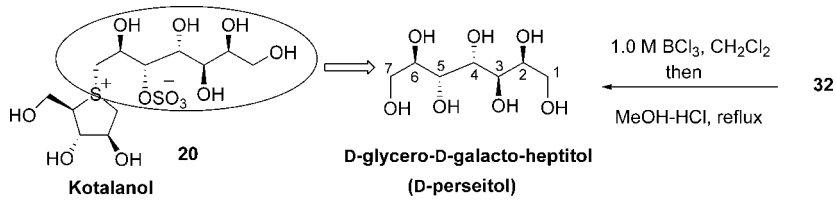
These results constitute, therefore, a formal structure proof of kotalanol **20**, the naturally occurring glycosidase inhibitor from *S. reticulata* (Scheme 6). It also confirms that the heptitol side chain of kotalanol is 5-*O*-sulfonyl-D-perseitol, a naturally occurring heptitol isolated from fruits,[23a] leaves,[23b] and the wound exudate[23c] of avocado trees. Further corroboration was obtained by comparison of the physical data of the heptitol, obtained upon treatment of intermediate **32** with 1.0 M BCl_3 (Scheme 6), with those of commercially available D-perseitol.

Having confirmed the structure of kotalanol, we embarked on its synthesis from a cyclic sulfate derived from D-perseitol. Thus, D-perseitol was first converted into 1,3:5,7-di-*O*-benzylidene-D-perseitol **41**,[24] and the secondary hydroxyl groups were then protected as PMB ethers. The protected compound was treated with a catalytic amount of PTSA in methanol to yield the regioisomeric diols, **42** and **43**, in 44% and 34% yield, respectively. These isomers were conveniently separated by column chromatography and differentiated by careful NMR analysis as shown in Scheme 7 and Table 3. Thus, the compound with lower coupling constant values for H-2 (J = 1.2, 1.2, 1.2 Hz) and showing HMBC correlations between the benzylidene acetal carbon and C-1 and C-3 was identified as being the desired diol **43**, to be taken on to the next step. The compound with higher coupling constants for H-6 (J = 4.8, 10.8, 9.6 Hz) and showing HMBC correlations between the benzylidene acetal carbon and C-5 and C-7 was identified as being the undesired diol **42**.

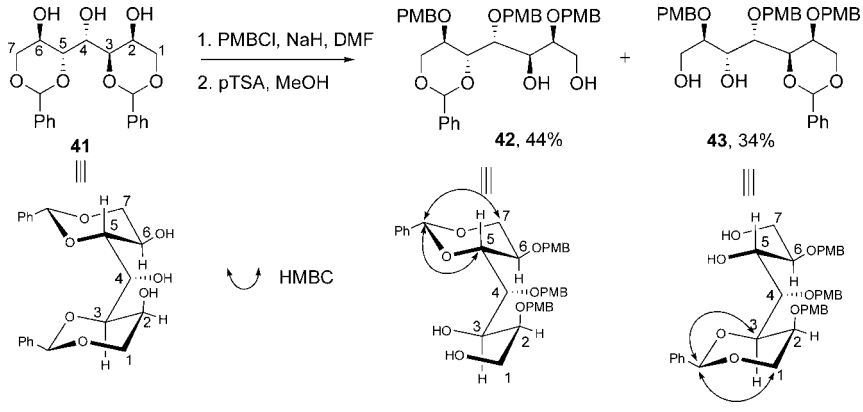
The desired diol **43** was first converted into the cyclic sulfate **44** and then coupled with the PMB-protected thio-D-arabinitol **35** as before to yield compound **45** in 69% yield. The PMB and benzylidene protecting groups were removed in one pot by treatment with 80% trifluoroacetic acid (TFA) in water at room temperature to yield compound **20** in 93% yield (Scheme 8).

Detailed 1D and 2D NMR experiments of compound **20** in pyridine-d_5 were performed, and the data were compared with those reported for kotalanol.[5] The choice of pyridine-d_5 as solvent caused broad peaks due to coupling with the hydroxyl groups, and hence, a D_2O exchange experiment was necessary to calculate the exact coupling constants (see Supporting Information). The stereochemistry at the stereogenic sulfur atom

Scheme 6



Scheme 7



Scheme 8

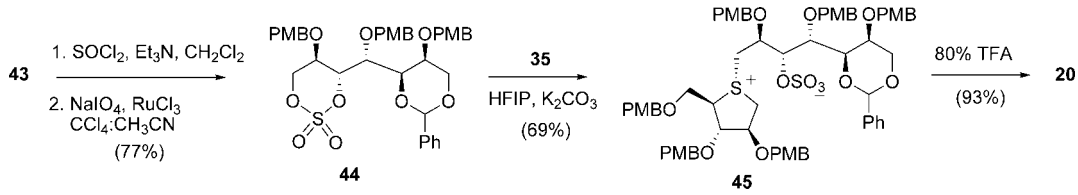


Table 4. Comparison of ^{1}H and ^{13}C NMR Data with Those Reported for Kotalanol in Pyridine-d_5

	^{1}H NMR data		^{13}C NMR data	
	20	kotalanol	**20**	kotalanol
1′	4.65, 4.93	4.65, 4.93	53.8	53.7
2′	5.24	5.24	67.4	67.4
3′	5.64	5.64	77.9	77.9
4′	5.12	5.12	70.5	70.5
5′	4.86	5.86	71.3	71.3
6′	4.88	4.88	72.6	72.5
7′	4.24,4.40	4.25, 4.50	65.4	65.3
1	4.31	4.31	50.1	50.2
2	5.07	5.08	78.1	78.1
3	5.15	5.16	79.4	79.4
4	4.62	4.64	72.2	72.2
5	4.51	4.51	60.0	60.0

was established by means of NOESY experiments in an analogous manner to those performed for the de-*O*-sulfonated compounds **38** and **40**. The ^{1}H NMR data of compound **20** in pyridine-d_5 compare favorably with those reported for kotalanol (with deviations of $\pm 0-0.1$ ppm), except for the chemical shift of H-5′ (Table 4). The reported chemical shift value for H-5′ was at 5.86 ppm,[5] whereas the ^{1}H NMR spectrum of compound **20** showed the corresponding signal at 4.86 ppm (confirmed with the aid of ^{1}H−^{1}H COSY, HMQC, and HMBC experiments). In fact, compound **20** had no signal appearing below 5.64 ppm. However, all of the ^{13}C NMR chemical shifts of compound **20** correlate well with those reported[5] for kotalanol, with deviations of $\pm 0-0.1$ ppm (Table 4). This mismatch of ^{1}H NMR chemical shift values of H-5′ was also one of the major discrepancies noted in our previously synthesized kotalanol analogues.[16] To eliminate this discrepancy unambiguously, we subjected compound **20** to de-*O*-sulfonation using the reported procedure[3] and compared the ^{1}H and ^{13}C NMR data (in CD_3OD) of the resulting de-*O*-sulfonated compound with those reported for de-*O*-sulfonated kotalanol.[3] These results indicated that, indeed, all ^{1}H and ^{13}C NMR chemical shift values agreed with those reported.[3] Hence, it is reasonable to conclude that the reported[5] chemical shift value for H-5′ in kotalanol must be in error. We note also that the optical rotation of compound **20** ($[\alpha]_D^{23}$ −5.7° (c = 0.7, MeOH)) is not in agreement with the reported value ($[\alpha]_D^{27}$ +11.5° (MeOH)); we obtained a specific rotation of +7.0° for **20** in water (c = 0.6, H_2O). To confirm the change in sign of optical rotation as a function of solvent, the optical rotation of the same sample was repeated in MeOH and in H_2O, alternately. Once again, in methanol, compound **20** showed levo (−) rotation and in water showed dextro (+) rotation. We attribute this discrepancy to the differential aggregation of this zwitterionic compound in MeOH and water. We note also that the optical rotations of the recent analogues of kotalanol, made by Muraoka et al.,[15b] were also reported in H_2O and that the reported data for kotalanol[5] do not indicate the concentration at which the optical rotation was measured. Hence, we surmise that the solvent reported[5] for the measurement of the optical rotation was also in error.

On the basis of the successful conversion of synthetic material **20** to de-*O*-sulfonated kotalanol and comparison of physical data with those of kotalanol (given the errors noted above), we conclude that the absolute stereostructure of kotalanol is the structure displayed by **20**, bearing the D-perseitol configuration in the acyclic side chain.

Experimental Section

See the Supporting Information for details.

Acknowledgment. We are grateful to the Canadian Institutes for Health Research for financial support. This article is dedicated, with respect and gratitude, to Professor T. B. Grindley on the occasion of his 65th birthday.

Supporting Information Available: Complete experimental section and ^{1}H and ^{13}C NMR spectra for compounds **20**, **25**−**34**, **37**, **38**, and **40**−**45**. This material is available free of charge via the Internet at http://pubs.acs.org.

JA900867Q

(24) Ness, A. T.; Hann, R. M.; Hudson, C. S. *J. Am. Chem. Soc.* **1948**, *70*, 765–770.