ORGANIC LETTERS

2010
Vol. 12, No. 7
1632−1635

Potent Glucosidase Inhibitors: De-*O*-sulfonated Ponkoranol and Its Stereoisomer

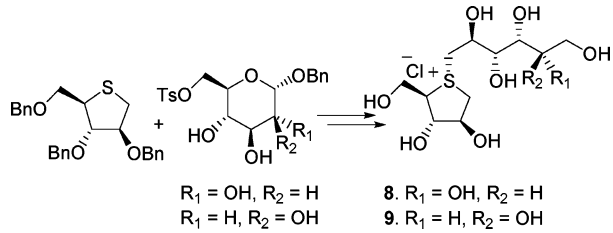
Razieh Eskandari,[†] Douglas A. Kuntz,[‡] David R. Rose,[‡,§] and B. Mario Pinto*[,†]

Department of Chemistry, Simon Fraser University, Burnaby, British Columbia, Canada V5A 1S6, Department of Medical Biophysics, University of Toronto and Division of Molecular and Structural Biology, Ontario Cancer Institute, Toronto, Ontario, Canada M5G 2M9, and Department of Biology, University of Waterloo, Waterloo, Ontario, Canada N2L 3G1

bpinto@sfu.ca

Received February 16, 2010

ABSTRACT



Ponkoranol, a glucosidase inhibitor isolated from the plant *Salacia reticulata*, comprises a sulfonium ion with an internal sulfate counterion. An efficient synthetic route to de-*O*-sulfonated ponkoranol and its 5′-stereoisomer is reported, and it is shown that these compounds are potent glucosidase inhibitors that inhibit a key intestinal human glucosidase, the *N*-terminal catalytic domain of maltase glucoamylase, with K_i values of 43 ± 3 and 15 ± 1 nM, respectively.

Compounds isolated from medicinal plants can provide the lead structures for drug development programs.[1,2] For example, the aqueous extracts of the roots and stems of the large woody climbing plant *Salacia reticulata,* known as Kothalahimbutu in Singhalese, have been used in the Ayurvedic system of Indian medicine in Sri Lanka and Southern India for the treatment of Type-2 diabetes.[3,4] Several glucosidase inhibitors have been isolated from the water-soluble fraction of this plant extract and also other plants that belong to the *Salacia* genus such as *Salacia chinensis*, *Salacia prinoides*, and *Salacia oblonga* which explain, at least in part, the antidiabetic property of the aqueous extracts of these plants.[5−7] Thus far, six components have been isolated from the plant *S. reticulata*, namely salaprinol (**1**),[7] salacinol (**2**),[6] ponkoranol (**3**),[7] kotalanol (**4**),[5] de-*O*-sulfonated kotalanol (**5**),[8] and de-*O*-sulfonated salacinol (**6**)[9] (Figure 1), all of which possess a common structural motif that comprises a 1,4-anhydro-4-thio-D-arabinitol and

[†] Simon Fraser University.
[‡] University of Toronto and Ontario Cancer Institute.
[§] University of Waterloo.

(1) Harvey, A. L. Natural products in drug discovery. *Drug Discov. Today* **2008**, *13*, 894–901.
(2) Butler, M. S. Natural products to drugs: natural product-derived compounds in clinical trials. *Nat. Prod. Rep.* **2008**, *25*, 475–516.
(3) Chandrasena, J. P. C. *The Chemistry and Pharmacology of Ceylon and Indian Medicinal Plants*; H&C Press: Colombo, Sri Lanka, 1935.
(4) Jayaweera, D. M. A. *Medicinal Plants Used in Ceylon-Part 1*; National Science Council of Sri Lanka: Colombo, 1981.
(5) Matsuda, H.; Li, Y. H.; Murakami, T.; Matsumura, N.; Yamahara, J.; Yoshikawa, M. *Chem. Pharm. Bull.* **1998**, *46*, 1399–1403.
(6) Yoshikawa, M.; Murakami, T.; Shimada, H.; Matsuda, H.; Yamahara, J.; Tanabe, G.; Muraoka, O. *Tetrahedron Lett.* **1997**, *38*, 8367–8370.
(7) Yoshikawa, M.; Xu, F. M.; Nakamura, S.; Wang, T.; Matsuda, H.; Tanabe, G.; Muraoka, O. *Heterocycles* **2008**, *75*, 1397–1405.
(8) Muraoka, O.; Xie, W. J.; Tanabe, G.; Amer, M. F. A.; Minematsu, T.; Yoshikawa, M. *Tetrahedron Lett.* **2008**, *49*, 7315–7317.
(9) Minami, Y.; Kurlyarna, C.; Ikeda, K.; Kato, A.; Takebayashi, K.; Adachi, I.; Fleet, G. W. J.; Kettawan, A.; Karnoto, T.; Asano, N. *Bioorg. Med. Chem.* **2008**, *16*, 2734–2740.

10.1021/ol1004005
Published on Web 03/10/2010

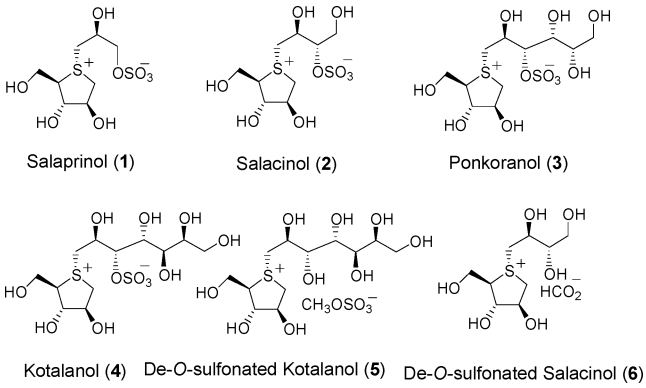
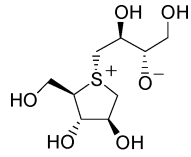


Figure 1. Components isolated from *Salacia* species.

a polyhydroxylated side chain. We have carried out extensive research on the synthesis of higher homologues of salacinol (**2**) which has led to the stereochemical structure elucidation of compounds **3**−**5**.[10−12] Interestingly, ponkoranol (**3**), the recently isolated[7] six-carbon-chain homologue of salacinol, was synthesized by us several years earlier with the expectation that it would be an effective glucosidase inhibitor.[13]

Recently, Minami et al.[9] reported the isolation of a thiosugar sulfonium−alkoxide inner salt (**7**), neosalacinol (Figure 2), from *S. reticulata*; however, Tanabe et al.[14] have

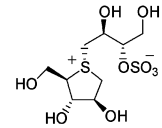
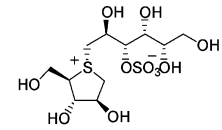
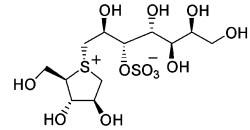
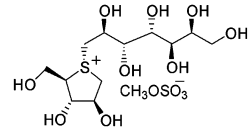
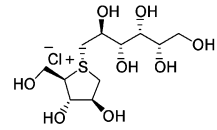
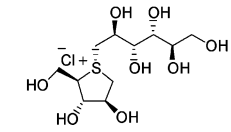


7

Figure 2. Proposed structure of neosalacinol.

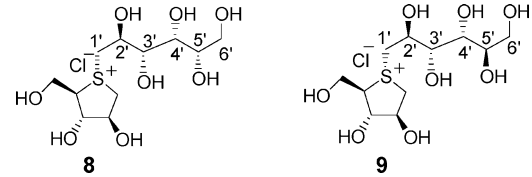
shown that this compound is de-*O*-sulfonated salacinol (**6**). Comparison of the inhibitory activities of de-*O*-sulfonated salacinol (**6**) vs salacinol (**2**) and de-*O*-sulfonated kotalanol (**5**) vs kotalanol (**4**) against rat intestinal α-glucosidases (maltase, sucrase, and isomaltase) revealed that the desulfonated analogues were either equivalent or better inhibitors than the parent compounds.[7,15,16] Furthermore, we have shown recently that de-*O*-sulfonated kotalanol (**5**) ($K_i = 0.03 \pm 0.01$ μM) is more potent an inhibitor of the *N*-terminal catalytic domain of human intestinal maltase glucoamylase (ntMGAM) than kotalanol (**4**) itself ($K_i = 0.19 \pm 0.03$ μM) (Table 1).[17]

Table 1. Experimentally Determined K_i Values[a]

Inhibitor		Ki (nM)
2		190 ± 20[19]
3		170 ± 30[19]
4		190 ± 30[17]
5		30 ± 10[17]
8		43 ± 1
9		15 ± 1

[a] Analysis of ntMGAM inhibition was performed using maltose as the substrate.

In view of these findings, it was of interest to question whether de-*O*-sulfonated ponkoranol **8** and its 5′-stereoisomer **9** (Figure 3) would be more potent inhibitors than ponkoranol itself.



8 **9**

Figure 3. De-*O*-sulfonated ponkoranol and its 5′-stereoisomer.

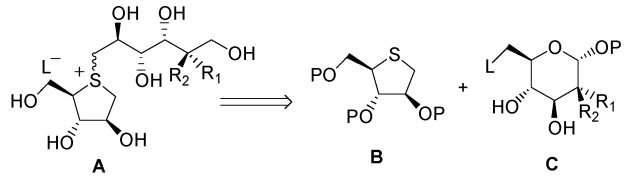
Our previous work with kotalanol analogues had suggested that the configuration at C-5′ was not critical for inhibitory

(10) Jayakanthan, K.; Mohan, S.; Pinto, B. M. *J. Am. Chem. Soc.* **2009**, *131*, 5621–5626.
(11) Mohan, S.; Pinto, B. M. *Carbohydr. Res.* **2007**, *342*, 1551–1580.
(12) Mohan, S.; Pinto, B. M. *Collect. Czech. Chem. Commun.* **2009**, *74*, 1117–1136.
(13) Johnston, B. D.; Jensen, H. H.; Pinto, B. M. *J. Org. Chem.* **2006**, *71*, 1111–1118.
(14) Tanabe, G.; Xie, W. J.; Ogawa, A.; Cao, C. N.; Minematsu, T.; Yoshikawa, M.; Muraoka, O. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 2195–2198.
(15) Matsuda, H.; Yoshikawa, M.; Murakami, T.; Tanabe, G.; Muraoka, O. *J. Trad. Med.* **2005**, *22*, 145–153.
(16) Ozaki, S.; Oae, H.; Kitamura, S. *J. Nat. Prod.* **2008**, *71*, 981–984.
(17) Sim, L.; Jayakanthan, K.; Mohan, S.; Nasi, R.; Johnston, B. D.; Pinto, B. M.; Rose, D. R. *Biochemistry* **2010**, *49*, 443–451.

activity.[17,18] We report here an efficient synthetic route to de-*O*-sulfonated ponkoranol **8** and its 5′-stereoisomer **9** and show that they are very potent inhibitors of the amino terminal catalytic domain of human maltase glucoamylase (ntMGAM).[19]

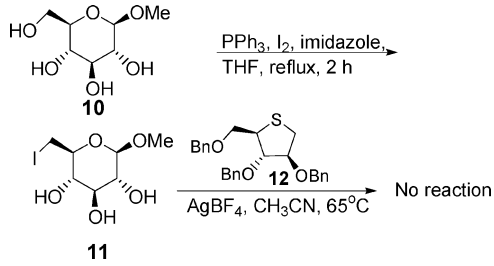
The sulfonium ions **A** could be synthesized by alkylation of an appropriately protected 1,4-anhydro-4-thio-D-arabinitol **B** at the ring sulfur atom with agent **C**. The desired stereochemistry at C-5′ could be readily obtained by choice of either D-glucose or D-mannose as starting material (Scheme 1).

Scheme 1. Retrosynthetic Analysis



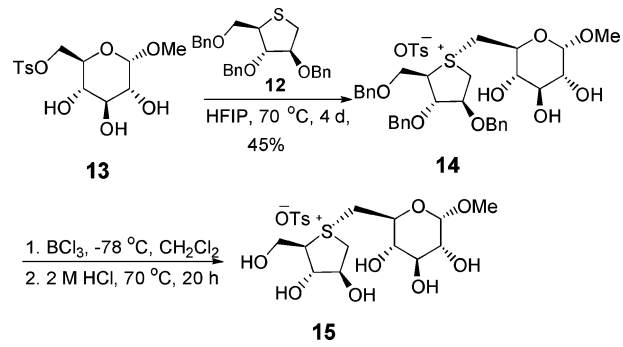
Initially, the S-alkylation of thioarabinitol **12**[20] with methyl 6-iodo-β-D-glucopyranoside **11**[21] in CH_3CN using $AgBF_4$ at 65 °C was examined, based on the procedure that has been reported for S-alkylation with simple alkyl halides (Scheme 2).[22] No product formation and decomposition of the starting

Scheme 2. First Attempted Synthesis of **8**



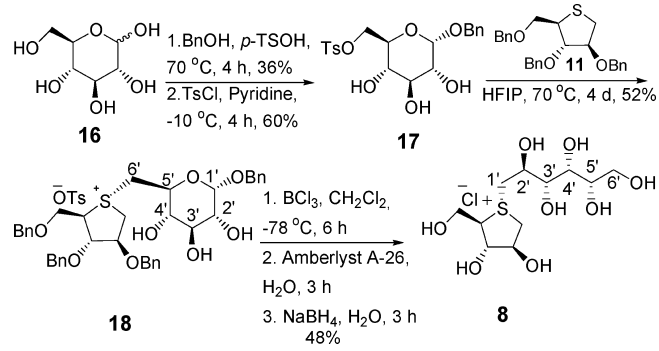
material were observed by TLC; the reaction in 1,1,1,3,3,3-hexafluoroisopropyl alcohol (HFIP)[23] as a solvent was also unsuccessful. In contrast, the coupling reaction with the *p*-toluenesulfonyl ester **13**[24] in HFIP at 70 °C proceeded smoothly and yielded the sulfonium ion **14** (Scheme 3). The

Scheme 3. Second Attempted Synthesis of **8**



benzyl groups of compound **14** were removed by treatment with boron trichloride at −78 °C in CH_2Cl_2. However, attempts to hydrolyze the methyl glycoside **15** with 2 M HCl were not successful, and decomposition of the product was observed. Therefore, a benzyl glycoside was chosen as a protecting group at the anomeric position to ensure its facile removal after the coupling reaction. Thus, benzyl 6-*O*-*p*-toluenesulfonyl-α-D-gluco **17** or mannopyranoside **20** were readily prepared from D-glucose and D-Mannose, respectively, according to literature procedures.[25−27] The thioether **12** was reacted with **17** in HFIP containing K_2CO_3[23] to give the protected sulfonium ion **18** in 52% yield (Scheme 4).

Scheme 4. Synthesis of Compound **8**

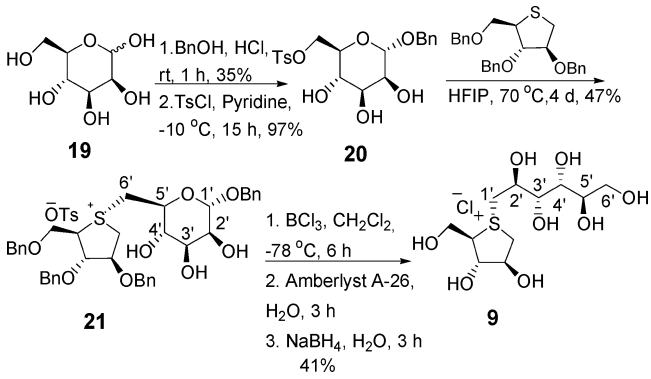


The benzyl groups were then removed by treatment with boron trichloride at −78 °C in CH_2Cl_2. During the course of deprotection, the *p*-toluenesulfonate counterion was

(18) Nasi, R.; Patrick, B. O.; Sim, L.; Rose, D. R.; Pinto, B. M. *J. Org. Chem.* **2008**, *73*, 6172–6181.
(19) Rossi, E. J.; Sim, L.; Kuntz, D. A.; Hahn, D.; Johnston, B. D.; Ghavami, A.; Szczepina, M. G.; Kumar, N. S.; Sterchi, E. E.; Nichols, B. L.; Pinto, B. M.; Rose, D. R. *FEBS J.* **2006**, *273*, 2673–2683.
(20) Satoh, H.; Yoshimura, Y.; Sakata, S.; Miura, S.; Machida, H.; Matsuda, A. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 989–992.
(21) Skaanderup, P. R.; Poulsen, C. S.; Hyldtoft, L.; Jorgensen, M. R.; Madsen, R. *Synthesis* **2002**, 1721–1727.
(22) Sankar, M.; Sim, L.; David, R. R.; Pinto, B. M. *Carbohydr. Res.* **2007**, *342*, 901–912.
(23) Ghavami, A.; Sadalapure, K. S.; Johnston, B. D.; Lobera, M.; Snider, B. B.; Pinto, B. M. *Synlett* **2003**, 1259–1262.
(24) Pellowska-Januszek, L.; Dmochowska, B.; Skorupa, E.; Chojnacki, J.; Wojnowski, W.; Wiśniewski, A. P. *Carbohydr. Res.* **2004**, *339*, 1537–1544.
(25) Andreana, P. R.; Sanders, T.; Janczuk, A.; Warrick, J. I.; Wang, P. G. *Tetrahedron Lett.* **2002**, *43*, 6525–6528.
(26) Branchaud, B. P.; Meier, M. S. *J. Org. Chem.* **1989**, *54*, 1320–1326.
(27) Koto, S.; Inada, S.; Yoshida, T.; Toyama, M.; Zen, S. *Can. J. Chem.* **1981**, *59*, 255–259.

partially exchanged with chloride ion. Similar results were observed in previous work from our laboratory.[22] Hence, after removal of the benzyl groups, the product was subsequently treated with Amberlyst A-26 resin (chloride form) to completely exchange the *p*-toluenesulfonate counterion with chloride ion. Finally, the crude product was reduced with $NaBH_4$ to provide the desired de-*O*-sulfonated ponkoranol **8** in 48% yield over three steps (Scheme 4). The other diastereomer was obtained similarly. Thus, compound **20** was reacted with the thioether **12** to give the protected sulfonium ion **21** in 47% yield which was converted, as before, to the desired compound **9** in 41% yield over three steps (Scheme 5).

Scheme 5. Synthesis of Compound **9**



The absolute stereochemistry at the stereogenic sulfur center in **18** and **21** was established by means of 1D-NOESY experiments (Figure 4) which showed H-4 to H-6′ correlations, implying that these atoms are syn-facial with respect to the sulfonium salt ring.

Finally, we comment on the inhibitory activities of compounds **8** and **9** against the *N*-terminus of recombinant human maltase glucoamylase (ntMGAM),[19] a critical intestinal glucosidase for postamylase processing of starch-derived oligosaccharides into glucose. The de-*O*-sulfonated ponkoranol **8** and its 5′-stereoisomer **9** inhibited ntMGAM with K_i values of 43 ± 3 and 15 ± 1 nM, respectively, both significantly lower than that (170 ± 30[19]) for ponkoranol (**3**) itself (Table 1). Thus, it would appear that de-*O*-sulfonation is beneficial. We have attributed this fact previously to alleviation of steric compression of the sulfate anion in a hydrophobic pocket within the active site of ntMGAM.[18] The K_i values for **8** and **9** compare to a K_i value for de-*O*-sulfonated kotalanol of 30 ± 1 nM.[17] It would appear, therefore, that the configuration at C-5′ is not critical for dictating enzyme inhibitory activity against ntMGAM and, furthermore, that extension of the acyclic carbon chain beyond six carbons is not essential. We note that **9** is the most potent compound to date in this class of molecules.

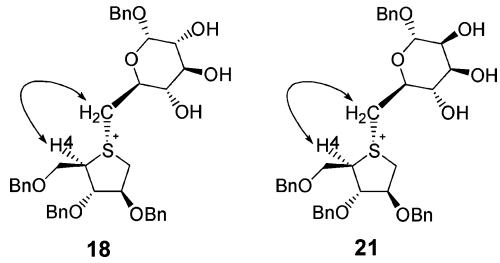


Figure 4. 1D-NOESY correlations of selected protons in compounds **18** and **21**.

Acknowledgment. We are grateful to the Canadian Institutes for Health Research (FRN79400) and the Heart and Stroke Foundation of Ontario (NA-6305) for financial support.

Supporting Information Available: Experimental procedures, characterization data, ^1H and ^{13}C NMR spectra of compounds **8**, **9**, **14**, **18**, and **21**, and 1D-NOESY spectra of compounds **18** and **21**. This material is available free of charge via the Internet at http://pubs.acs.org.

OL1004005