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Synthesis and Biological Activities of Inositol 1, 4, 5-Trisphosphate Mimics Related to Xylopyranosides

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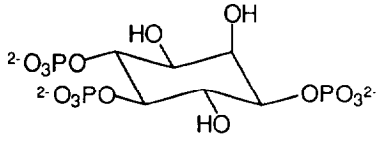
Abstract: *2', 3, 4-trisphosphates of (2-hydroxyethyl) α- and β-D-xylopyranosides and 3', 3, 4-trisphosphates of (3-hydroxypropyl) α– and β-D-xylopyranosides have been prepared from allyl-D-xylosides and showed agonistic properties toward inositol 1, 4, 5-trisphosphate receptor.*

The discovery of inositol-1, 4, 5-trisphosphate ($InsP_3$) **1** as a second messenger [1,2] in response to extracellular messages has prompted numerous studies in the synthesis of analogues of $InsP_3$ [3,4]. Until recently only close analogues to $InsP_3$ have been reported, including modifications of the substitution pattern of hydroxyl groups of inositols or modifications of the phosphate moieties.[5] Considering the weak influence, if any, of the 2- and 3-OH groups,[6] we tried to devise new analogues of $InsP_3$ which should be more accessible. Given the good availability of sugars, we reasoned that they may act as surrogates of the inositol ring lacking the 2- and 3-OH groups.[7] In the event, D-xylose derivatives appeared to be good candidates. In particular β-D-xylopyranose-1, 3, 4-trisphosphate was very attractive. We embarked in the synthesis of this derivative which proved to be rather unstable. Thus we planned to replace the anomeric phosphate by an hydroxyalkyl chain which could be further phosphorylated.

In the meantime, the discovery of adenophostin A **2** and related compounds, which are potent agonists of $InsP_3$ receptor with high mobilizing calcium activity shed an interesting light on our synthetic plan.[8] More recently, Potter and Jenkins reported the synthesis of second messenger mimic related to adenosphostin A.[9] This prompted us to report in this letter our results in the synthesis of four $InsP_3$ mimics which are in turn also adenophostins mimics. Preliminary results of their biological activities are also reported.



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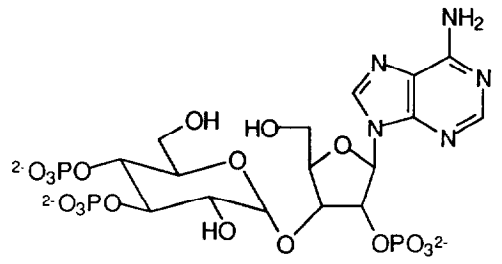

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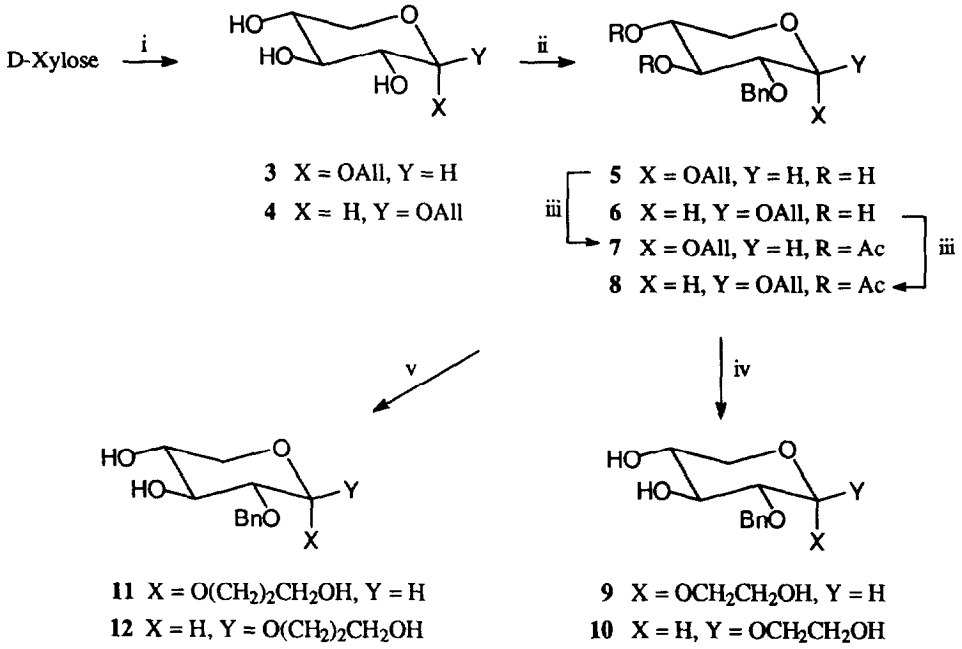
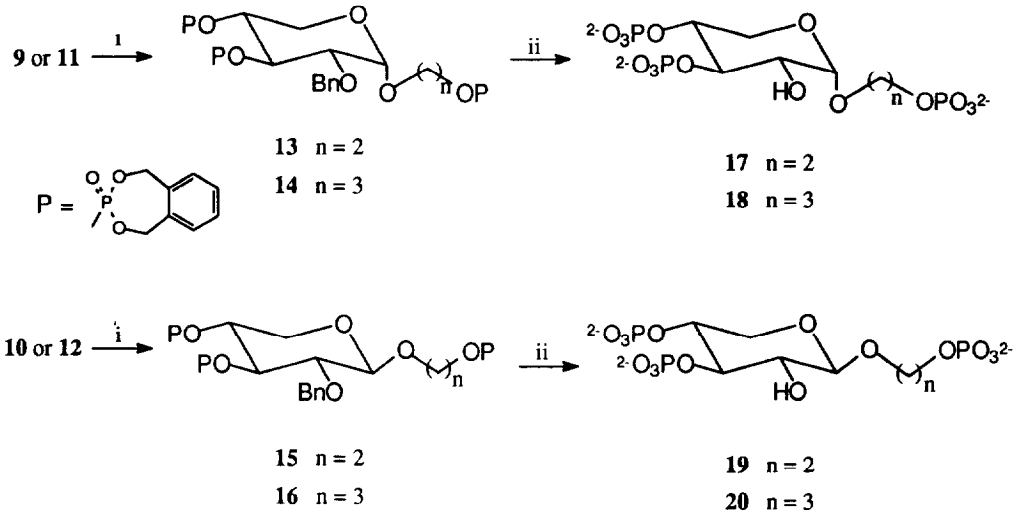
D-xylose was treated with allyl alcohol under Fischer conditions to give the allyl glycosides **3** and **4** as a 2:1 α/β mixture. Acetylation of this mixture gave the corresponding allyl-α and β-tri-*O*-acetylxylopyranosides which were separated by preparative high pressure column chromatography. Each compound was deacetylated to give **3** and **4** in 30 and 16% yields respectively. Selective benzylation at position 2 was attempted on each compounds. In the case of the α derivative **3** the stannylene method was used and provided the desired 2-*O*-benzyl derivative **5** in 40% yield.[10] However, the β derivative **4** cannot be alkylated efficiently by the same method. Nevertheless, mono benzylation can be performed by conventional method (NaH, BnBr) although with a poor selectivity. Careful chromatography of the mixture gave the 2-OBn β derivative **6** in 20% yield. Standard acetylation of compound **5** and **6** provided fully protected **7** and **8** in quantitative yields.



Scheme 1. Reagents and conditions: **i:** a)AllOH / Dowex 50W H^+, reflux, 48 h; b) Ac_2O / pyridine ; c) MeOH / MeONa. **ii:** a)Bu_2SnO, toluene, reflux ; b) BnBr, 60°C, 7h for **5** or BnBr, DMF, 0°C for **6**; **iii:** Ac_2O / pyridine; **iv:** a) O_3, MeOH / THF, 1h 30; -70°C; b) $NaBH_4$; c) MeOH, MeONa; **v:** a) 9-BBN, THF, ultrasonic irradiation, 1h 30; b) NaOH, H_2O_2; c) MeOH, MeONa.

Introduction of the required hydroxymethyl functionalities by chemical manipulation of the aglycon was then examined. Ozonolysis of **7** or **8** followed by reductive work-up with sodium borohydride gave the expected 2-hydroxyethyl-xylosides **9** and **10** respectively. The corresponding 3-hydroxy propyl derivatives **11** and **12** were obtained by hydroboration of the double bond. The reaction performed with 9-BBN in tetrahydrofuran gave the best results provided ultrasonic irradiation was used.[11] Upon oxidative treatment, alcohols **11** and **12** were obtained in 51 and 61% yields respectively.

Phosphitylation of the four triols **9**, **10**, **11** and **12** was performed by reaction with excess of 2-(N,N-diisopropylamino)-5,6-benzo-1,3,2-dioxaphosphepane in the presence of tetrazole.[12] The phosphites were subsequently oxidized with *t*-butyl peroxide. Careful purification provided analytically pure protected trisphosphates **13**, **14**, **15** and **16** from **9**, **11**, **10** and **12** respectively. Spectroscopic data ^{1}H, ^{13}C and ^{31}P nmr were consistent with the expected structures. Hydrogenolysis of the benzyl group gave the free phosphates **17**, **18**, **19**, **20** respectively from which the corresponding hexakis sodium salts were prepared by neutralisation with sodium hydroxide.



Scheme 2: Reagents and conditions : **i** a) 2-(N,N-diisopropylamino)-5,6-benzo-1,3,2-dioxaphosphepane, tetrazole, CH_2Cl_2, 3h, rt: b) tBuOOH; **ii** H_2 Pd/C 10% 50 psi.

The $InsP_3$ receptor binding properties of **17**, **18**, **19**, and **20** were assayed on rat cerebellar membranes. All these compounds are full agonists of $InsP_3$ with EC_{50} of 0.43, 0.78, 1.7, and 4.8 μM respectively. The ability of these compounds to release Ca^{2+} from permeabilized hepatocytes has been tested. The four compounds released Ca^{2+} from the intracellular compartment at around 10 μM concentration in the following order **17** ≅ **19** ≅ **18** > **20**. Their potency is roughly ten-fold lower than that of $InsP_3$. These results are in agreement with the fact that the 2- and 3-OH groups do not play an important role in the binding of $InsP_3$ to its receptor and thus the cyclitol moiety could be replaced by a pyranose ring. Moreover in the context of the adenophostins mimics it might be concluded, as expected, that the phosphates at C-2', C-3" and C-4" are essential for the biological activities[13] but that another functional group should be responsible for the high potency of adenophostins which release calcium at nM concentrations (100-fold more potent than $InsP_3$).[8c] It is reasonable to think that, rather than the hydroxymethyl group at C-5", the adenine group might play an important role in the binding. Another reason for the lower activities of our mimics compared to adenophostins could be the flexibility of the alkyl chain carrying the primary phosphate. We are currently investigating these hypotheses by synthetizing new analogues of **17** incorporating the purine moiety and conformationally restricted analogues.

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References and Notes

1. Berridge, M. J.; Irvine, R. F. *Nature*, **1984**, *312*, 315-321.
2. Berridge, M. J.; Irvine, R. F. *Nature*, **1989**, *341*, 197-203.
3. Billington, D. C. *The Inositol Phosphates: Chemical Synthesis and Biological Significance*, VCH, Weinheim, **1993**, pp 1-153.
4. Potter, B. V. L.; Nahorski, S. R. Synthetic inositol polyphosphates and analogues as molecular probes for neuronal second messengers receptors in "*Drug Design for Neuroscience*", Raven Press, New York, **1993**, Vol. 14, pp 383-416.
5. (a) Schmitt, L.; Spiess, B.; Schlewer, G. *Tetrahedron Lett.*, **1992**, *33*, 2013-2016; (b) Wilcox, R. A.; Nahorski, S. R.; Sawyer, D. A.; Liu, C.; Potter, B. V. L. *Carbohydr. Res.*, **1992**, *234*, 237-246; (c) Lampe, D.; Potter, B. V. L. *Tetrahedron Lett.*, **1993**, *34*, 2365-2368; (d) Kozikowski, A. P.; Ognyanov, V. I.; Fauq, A. H.; Wilcox, R. A.; Nahorski, S. R. *J. Chem. Soc., Chem. Commun.*, **1994**, 599-600; (e) Poirot, E.; Bourdon, H.; Chrétien, F.; Chapleur, Y.; Berton, B.; Hilly, M.; Mauger, J. -P.; Guillon, G. *Bioorg. Med. Chem. Lett.*, **1995**, *5*, 569-572 and references cited in these papers.
6. Kozikowski, A. P.; Ognyanov, V. I.; Fauq, A. H.; Nahorski, S. R.; Wilcox, R. A. *J. Am. Chem. Soc.*, **1993**, *115*, 4429-4434.
7. Riley, A. M.; Jenkins, D. J.; Potter, B. V. L. *J. Am. Chem. Soc.*, **1995**, *117*, 3300-3301.
8. (a) Takahashi, M.; Kagasaki, T.; Hosoya, T.; Takahashi, S. *J. Antibiotics*, **1993**, *46*, 1643-1647; (b) Takahashi, S.; Kinoshita, T.; Takahashi, M. *J. Antibiotics*, **1994**, *47*, 95-100; (c) Takahashi, M.; Tanzawa, K.; Takahashi, S. *J. Biol. Chem.*, **1994**, *269*, 369-372; (d) Hotoda, H.; Takahashi, M.; Tanzawa, K.; Takahashi, S.; Kaneko, M. *Tetrahedron Lett.*, **1995**, *36*, 5037-5040.
9. Jenkins, D. J.; Potter, B. V. L. *J. Chem. Soc., Chem. Commun.*, **1995**, 1169-1170. Another interesting approach to adenophostins mimics from D-galactose has been recently disclosed: Desai, T.; J. Gigg, J.; Gigg, R. VIII Eurocarb, Seville, Spain, 2-8 July 1995, communication A-14.
10. (a) Qin, H.; Grindley, T. B. *J. Carbohydr. Chem.*, **1994**, *13*, 475-486; (b) Helm, R. F.; Ralph, J.; Anderson, L. *J. Org. Chem.* **1991**, *56*, 7015-7021.
11. For the use of ultrasonic activation for hydroboration reactions see for example: Keck, G. E.; Palani, A. McHardy, S. F. *J. Org. Chem.* **1994**, *59*, 3113.
12. Uhlmann, E.; Engel, J. *Tetrahedron Lett*, **1986**, *41*, 1023-1026.
13. It is interesting to note that the more potent compounds of our series are the α derivatives **17** and **18** and in particular **17** which has a two carbon chain as in adenophostins.

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