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SYNTHESIS OF 2-FLUORO-2-DEOXY-*MYO*-INOSITOL 1,4,5-TRISPHOSPHATE AND *SCYLLO*-INOSITOL 1,2,4-TRISPHOSPHATE, NOVEL ANALOGUES OF THE SECOND MESSENGER *MYO*-INOSITOL 1,4,5-TRISPHOSPHATE

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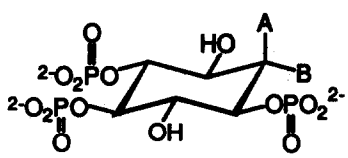
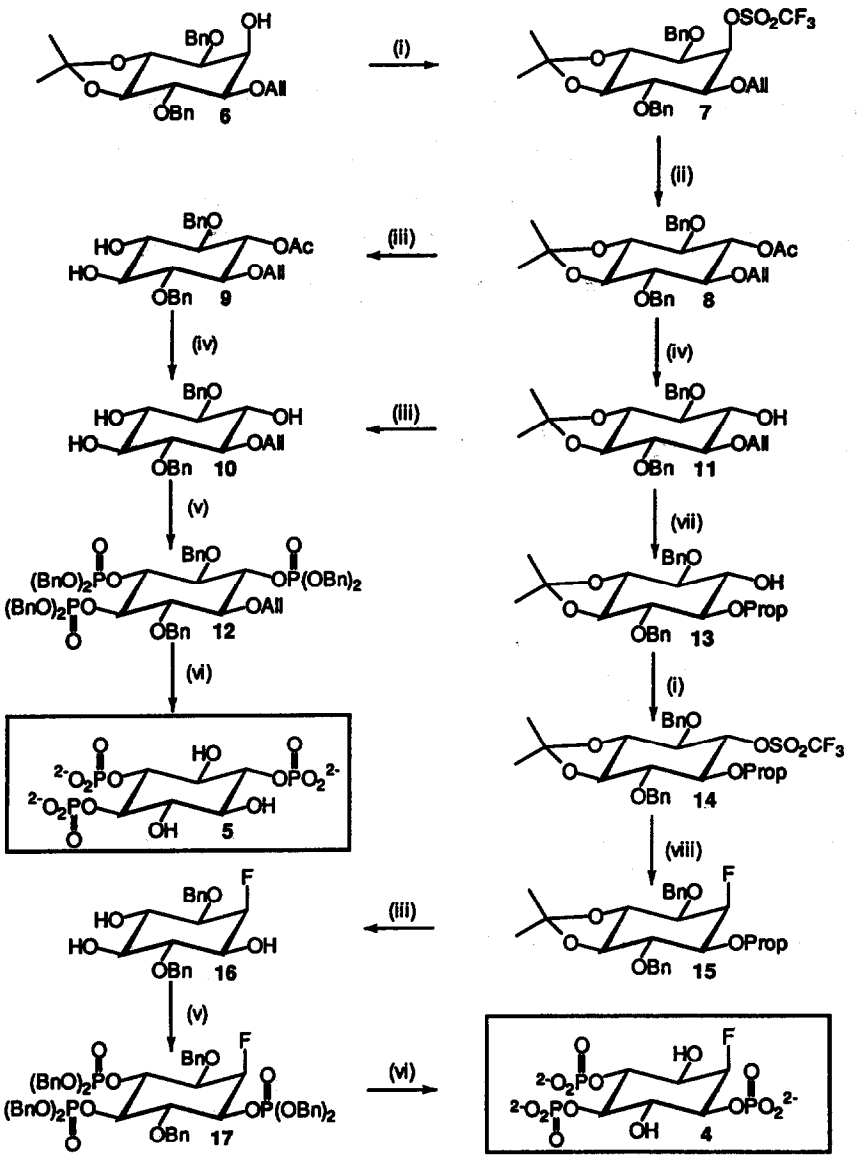
Abstract: *The novel* myo-*inositol 1,4,5-trisphosphate receptor agonists, 2-fluoro-2-deoxy-*myo-*inositol 1,4,5-trisphosphate and* scyllo-*inositol 1,2,4-trisphosphate, were synthesised in racemic form from suitably protected inositol precursors.*

D-*myo*-inositol 1,4,5-trisphosphate [(1), Figure] is a second messenger which releases Ca^{2+} from intracellular stores[1,2] via an isolated[3], cloned[4] and sequenced[5] receptor which when reconstituted, mediates Ca^{2+} release in response to Ins(1,4,5)P_3[6]. A major challenge is now the eludication of the structural basis for interaction of Ins(1,4,5)P_3 with its receptor and the metabolic enzymes, Ins(1,4,5)P_3 3-kinase and 5-phosphatase, and the rational chemical design of agonists, antagonists and enzyme inhibitors. Recent progress in inositol phosphate chemistry[7,8] and molecular recognition has been reviewed[9].



(1) A = OH, B = H
(2) A = H, B = F
(3) A = B = F
(4) A = F, B = H
(5) A = H, B = OH

Ring- and phosphate-modified analogues have been synthesized[7,8]. Isosteric replacement of a hydroxyl group with fluorine has led to fluorinated *myo*-inositol analogues[10-17], derivatives[18], inositol phosphate analogues[19-24] and lipids including 2-fluoro-2-deoxy-1-phosphatidyl-*scyllo*-inositol[25] and 3-fluoro-3-deoxyphosphatidylinositol[26]. D-3-fluoro-3-deoxy-*myo*-inositol inhibits cell growth[13] and 5-fluoro-5-deoxy-*myo*-inositol is incorporated into phospholipid by PtdIns synthase[27], although 5,5-difluoro-5-deoxy-*myo*-inositol is a much poorer substrate[15]. We reported the synthesis[28] and biological evaluation of the fluorinated inositol phosphate analogues, 2-fluoro-2-deoxy-*scyllo*-inositol 1,4,5-trisphosphate (2)[22], 2,2-difluoro-2-deoxy-Ins(1,4,5)P_3 (3)[22] and 3-fluoro-3-deoxy Ins(1,4,5)P_3[29] with the Ca^{2+}-releasing receptor



Scheme: *Reagents and conditions* i, $(CF_3SO_2)_2O$ (1.75 equiv.) - pyridine-CH_2Cl_2, 0°C; ii, cesium acetate (1.5 equiv.) - DMF; iii, MeOH-1 M HCl (5:1, v/v), reflux; iv, MeOH-1M NaOH (5:1, v/v), reflux; v, (a) $(BnO)_2PNPr^i_2$ (9 equiv.) - tetrazole (12 equiv.) in CH_2Cl_2, (b) Bu^tOOH (70% in H_2O); vi, Na-liq. NH_3; vii, Bu^tOOK-DMSO, 50°C; viii, Bu_4NF-THF. All compounds are racemic.

and enzymes 5-phosphatase and 3-kinase. L-2,2-difluoro-2-deoxy-Ins(1,4,5)P_3 is a potent inhibitor of 3-kinase and 5-phosphatase[30].

While the value of fluorinated derivatives of Ins(1,4,5)P_3 has clearly been demonstrated, it has not yet proved possible to synthesise what is arguably the most important 2-fluorinated Ins(1,4,5)P_3 analogue, 2-fluoro-2-deoxy Ins(1,4,5)P_3 (**4**), which possesses an axial 2-fluorine atom. An attempt was made to synthesise the corresponding fluorinated Ins(1,3,4)P_3 analogue[20], but was unsuccessful as a result of decomposition of the intermediate during deblocking. Fluorination of a protected *scyllo*-inositol precursor in an approach to (**4**) gave unexpectedly the product of retention of configuration[19]. We now report the successful synthesis of (**4**) and a related novel analogue *scyllo*-inositol 1,2,4-trisphosphate (**5**)[31], where the unique axial hydroxyl group of (**1**) has been inverted.

1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene *myo*-inositol (**6**)[32] (Scheme) was converted to the corresponding triflate (**7**)[33] (yield quantitative by tlc, compound not isolated) and the 2-position was inverted by reaction of (**7**) with cesium acetate to give (**8**) (yield 91%). The isopropylidene group of (**8**) was removed with acid and the acetate (**9**) saponified to yield the triol (**10**) (yields 83% and 87%, respectively). Phosphitylation of (**10**) with N,N-diisopropyldibenzyl phosphoramidite-tetrazole[34] followed by oxidation of the resulting trisphosphite gave the fully protected trisphosphate (**12**) (yield 58%). Treatment of (**12**) with sodium in liquid ammonia removed all protecting groups including the allyl group[35] to give trisphosphate (**5**), which was purified by ion-exchange chromatography on Q Sephadex Fast Flow using a gradient of triethylammonium bicarbonate as eluant (yield 91%).

In order to synthesise (**4**), acetate (**8**) was saponified and the allyl group isomerised[36] to give (**13**) (yields 85% and 95%), which was converted to the corresponding triflate (**14**). Inversion of configuration at C-2 by displacement of triflate using tetrabutylammonium fluoride gave (**15**) (yield 69%). Observation of $^3J_{HF}$ = 29.5 Hz in the NMR spectrum of (**15**) confirmed the assignment of an *axial* 2-fluorine atom. Removal of the isopropylidene and prop-1-enyl group generated triol (**16**) (yield 69%). Phosphorylation of (**16**) (yield 76%) and deblocking of the protected trisphosphate (**17**) as above gave (**4**) (yield 41%) which was purified by ion-exchange chromatography[37].

Racemic (**4**) and (**5**) bound to the Ins(1,4,5)P_3 receptor with high affinity and were Ca^{2+}-mobilising agonists with potencies very similar to Ins(1,4,5)P_3 itself[38]. These compounds will be useful in exploring structure-activity relationships and molecular recognition at Ins(1,4,5)P_3 binding proteins. Biological results will be reported elsewhere.

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REFERENCES AND NOTES

1. Berridge, M.J., *Annu. Rev. Biochem.*, **1987**, *56*, 159-193.
2. Berridge, M.J.; Irvine, R.F., *Nature*, **1989**, *341*, 197-205.
3. Supattapone, S.; Worley, P.F.; Baraban, J.M.; Snyder, S.H., *J. Biol. Chem.*, **1988**, *263*, 1530-1534.
4. Furuichi, T.; Yoshikawa, S.; Miyawaki, A.; Wada, K.; Maeda, N.; Mikoshiba, K., *Nature*, **1989**, *342*, 32-38.
5. Mignery, G.A.; Newton, C.L.; Archer III, B.T.; Südhof, T.C., *J. Biol. Chem.*, **1990**, *265*, 12679-12685.
6. Ferris, C.D.; Huganir, R.L.; Supattapone, S.; Snyder, S.H., *Nature*, **1989**, *342*, 87-89.
7. Billington, D.C., *Chem. Soc. Rev.*, **1989**, *18*, 83-122.
8. Potter, B.V.L., *Nat. Prod. Reports*, **1990**, *7*, 1-24.
9. Nahorski, S.R.; Potter, B.V.L., *Trends Pharmacol. Sci.*, **1989**, *10*, 139-144.
10. Lowe, G.; McPhee, F., *J. Chem. Soc. Perkin Trans. I*, **1991**, 1249-1253.
11. Kozikowski, A.P.; Xia, Y.; Rusnak, J.M., *J. Chem. Soc. Chem. Commun.*, **1988**, 1301-1303.
12. Kozikowski, A.P.; Fauq, A.H.; Rusnak, J.M., *Tetrahedron Lett.*, **1989**, *30*, 3365-3368.
13. Kozikowski, A.P.; Fauq, A.H.; Powis, G.; Melder, D.C., *J. Am. Chem. Soc.*, **1990**, *112*, 4528-4531.
14. Jiang,C.; Moyer, J.D.; Baker, D.C., *J. Carbohydr. Chem.*, **1987**, *6*, 319-355.
15. Jiang, C.; Schedler, D.J.A.; Morris Jr., P.E.; Zayed, A.-H.A.; Baker, D.C., *Carbohydr. Res.*, **1990**, *207*, 277-285.
16. Offer, J.L.; Metcalfe, J.C.; Smith, G.A., *J. Chem. Soc. Chem. Commun.*, **1990**, 1312-1313.
17. Yang, S.S.; Beattie, T.R.; Shen, T.Y., *Synth. Commun.*, **1986**, *16*, 131-138.
18. Yang, S.S.; Min, J.M.; Beattie, T.R., *Synth. Commun.*, **1988**, *18*, 899-905.
19. Marecek, J.F.; Prestwich, G.D., *Tetrahedron Lett.*, **1989**, *30*, 5401-5405.
20. Boehm, M.F.; Prestwich, G.D., *Tetrahedron Lett.*, **1988**, *29*, 5217-5220.
21. Ley, S.V.; Parra, M.; Redgrave, A.J.; Sternfield, F.; Vidal, A., *Tetrahedron Lett.*, **1989**, *30*, 3557-3560.
22. Safrany, S.T.; Sawyer, D.; Wojcikiewicz, R.J.H.; Nahorski, S.R.; Potter, B.V.L., *FEBS Lett.*, **1990**, *276*, 91-94.
23. Kozikowski, A.P.; Fauq, A.H.; Aksoy, I.A.; Seewald, M.J.; Powis, G., *J. Am. Chem. Soc.*, **1990**, *112*, 7403-7404.
24. Carless, H.A.J.; Busia, K., *Carbohydr. Res.*, **1992**, *234*, 207-215.
25. Yang, S.S.; Beattie, T.R.; Shen, T.Y., *Tetrahedron Lett.*, **1982**, *23*, 5517-5520.
26. Kozikowski, A.P.; Tückmantel, W.; Powis, G., *Angew. Chem.*, **1992**, *104*, 1408- 1410; *Angew. Chem. Int. Ed. Eng.*, *31*, 1381-1383.
27. Moyer, J.D.; Reizes, O.; Surender, A.; Jiang, C.; Malinowski, N.; Baker, D.C. *Mol. Pharmacol.*, **1988**, *33*, 683-689.
28. Sawyer, D.; Potter, B.V.L., *J. Chem. Soc. Perkin Trans. I*, **1992**, 923-932.
29. Safrany, S.T.; Liu, C.; Potter, B.V.L.; Nahorski, S.R., *Eur. J. Pharmacol.*, **1992**, *226*, 265-272.
30. Safrany, S.T.; Sawyer, D.; Nahorski, S.R.; Potter, B.V.L., *Chirality*, **1992**, *4*, 415-422.
31. Note that the structures of (**5**) in the Figure and Scheme are identical.
32. Gigg, J.; Gigg, R.; Payne, S.; Conant, R., *J. Chem. Soc. Perkin Trans. I*, **1987**, 1757-1762.
33. All new compounds exhibited satisfactory spectroscopic and microanalytical data.
34. Yu, K.-L.; Fraser-Reid, B., *Tetrahedron Lett.*, **1988**, *29*, 979-982.
35. Baldwin, S.W.; Tomesch, J.C., *J. Org. Chem.*, **1974**, *39*, 2382-2385.
36. Price, C.C.; Snyder, W.H., *J. Am. Chem. Soc.*, **1961**, *83*, 1773.
37. NMR data for (**4**). δ_H (270 MHz; D_2O) 3.86 (1 H, ddd, J 28.5, 9.9, 2.0, C-3-H), 3.90 (1 H, t, J 9.5, C-6-H), 4.09 (1 H, q, J 9.2, C-4(5)-H), 4.14 (ddt, J 27.5, 8.4, 1.7, C-1-H), 4.29 (1 H, q, J 9.2, C-5(4)-H), 5.10 (1 H, dt, J 51.8, 1.5, C-2-H); δ_P (36 MHz) 1.96 (1 P, J 10.1), 1.56 (1 P, J 6.7), 0.37 (1 P, J 10.1).
38. Compounds were evaluated in competitive receptor binding assays against [^{3}H]-Ins(1,4,5)P_3 and as agonists in $^{45}Ca^{2+}$ release assays in permeabilised SH-SY5Y neuroblastoma cells; see references 22, 29.

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