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Expedient Synthesis of D - myo - Inositol 1,4,5 - Trisphosphate and D - myo - Inositol 1,4 - Bisphosphate

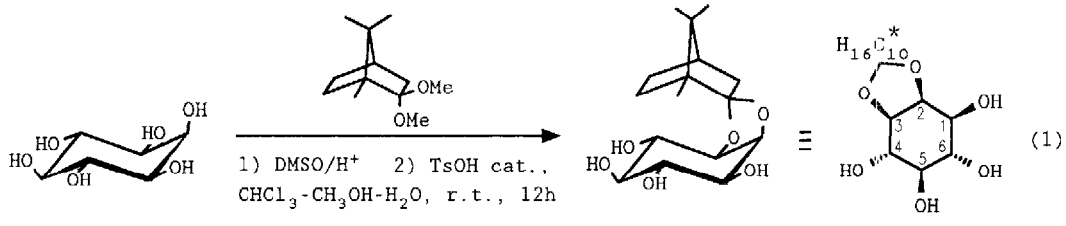
Grzegorz M. Salamończyk and K. Michał Pietrusiewicz*

Centre of Molecular and Macromolecular Studies, The Polish Academy of Sciences, Sienkiewicza 112, 90-363 Łódź, Poland

Key words: D-*myo*-inositol monoacetal, camphor, selectivity, acylation, silylation, phosphitylation.

Abstract: Readily available selfresolving *myo*-inositol D-camphor 2,3-monoacetal is converted into the title inositol phosphates by the concise procedures utilizing 1,4,5-selective tris-acylation and 1,4-selective bis-silylation of the starting tetrol in the key steps.

It is now well established that D-*myo*-inositol 1,4,5-trisphosphate [Ins(1,4,5)P_3], produced in cells by the receptor-controlled hydrolysis of phosphatidylinositol 4,5-bisphosphate [(Ptd)Ins(4,5)P_2], acts as a second messenger and stimulates the release of calcium from intracellular stores.[1] According to one of the known metabolic cycles[1] the action of Ins(1,4,5)P_3 is terminated by a sequence of specific monodephosphorylations leading *via* 1,4-bisphosphate [Ins(1,4)P_2], and 4-monophosphate to free inositol which is subsequently recycled in the resynthesis of (Ptd)Ins(4,5)P_2.[2] In this communication we wish to present an efficient synthetic route to homochiral Ins(1,4,5)P_3[3] and Ins(1,4)P_2.[4] The route commences with a recently introduced selfresolving 2,3-*O*-(D-1,7,7-trimethyl[2.2.1]bicyclohept-2-ylidene)-*myo*-inositol(**1**)[5] which can be expeditiously prepared from the parent cyclitol and D-camphor dimethyl acetal by way of the precipitation-driven equilibration depicted in Equation 1.[6]



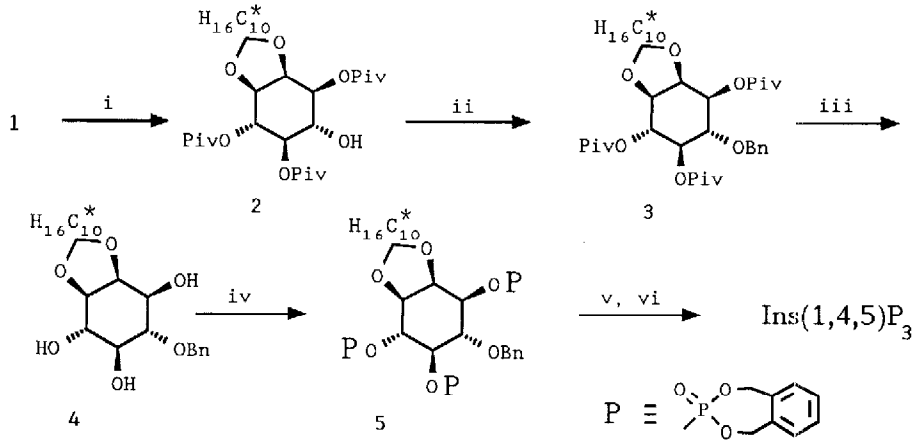
myo-inositol

(1)

1

As indicated already by the successful monophosphorylation of 1[6] and confirmed now more systematically in a series of exploratory experiments the four hydroxy groups in 1 can be effectively distinguished between chemically. Thus, treatment of 1 with 1.1 mol. equiv. of either pivaloyl chloride in pyridine or *tert*-butylchlorodiphenylsilane in the presence of imidazole in acetonitrile solution leads to the formation of the corresponding C1-OH protected derivative in 45 and 34% isolated yield, respectively,in full concordance with the phosphorylation results.[6] In contrast, when 1 is allowed to react with 2.2 mol. equiv. of the above reagents a less uniform picture obtains. Bis-acylation of 1 with pivaloyl chloride gives the corresponding 1,5-protected derivative in 43% yield, whereas bis-silylation with *tert*-butylchlorodimethylsilane affords the corresponding 1,4-protected derivative in comparable 50% yield.[7] In turn, when 1 is treated with an excess of pivaloyl chloride (5 mol. equiv.), the protection of only three hydroxy groups takes place and provides the 1,4,5-trispivaloyl derivative in 48% yield. Based on these exploratory experiments are the preparations of the title inositol phosphates which follow[8] (Scheme 1 and 2).

Scheme 1.

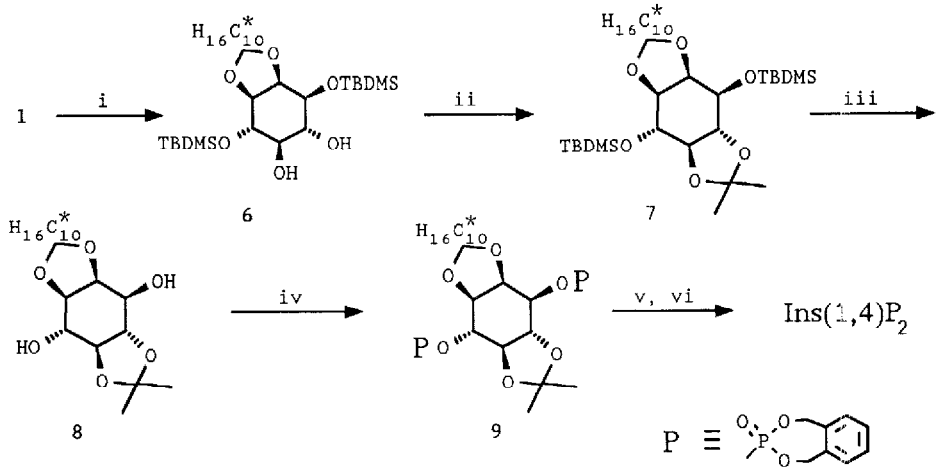


(i)$(CH_3)_3CC(O)Cl$, pyridine; (ii)BnBr, Ag_2O, DMF; (iii)NaOH, MeOH; (iv)a: 2-dimethylamino-5,6-benzo-1,3,2-dioxaphosphepane, CH_2Cl_2; b:MCPBA; (v) H_2, Pd/C, methanol; (vi)AcOH-H_2O(1:1)

As mentioned above, treatment of 1 with an excess of pivaloyl chloride in pyridine solution (5 mol. equiv.,r.t.,16 h), gave the trispivaloyl derivative 2 (Scheme 1), m.p. 110-112°C, $[\alpha]_D$=-12° (c 2.1, $CHCl_3$), in 48% isolated yield. Subsequent treatment of 2 with benzyl bromide (6 mol. equiv.) and silver(I) oxide (3 mol. equiv.) at 4°C for 24h afforded fully protected inositol 3, m.p. 184-185°C, $[\alpha]_D$=-14° (c 2.1, $CHCl_3$), in 50% yield. After deblocking of 3 with NaOH in refluxing CH_3OH for 1h, the resulting triol 4, m.p. 44-45°C, $[\alpha]_D$=+16.2° (c 2.5, $CHCl_3$), 92%, was subjected to phosphorylation using a slight modification of the procedure reported by Watanabe *et al.*[9] Thus, 4 was allowed to react with 4.5 mol.

equiv. of 2-dimethylamino-5,6-benzo-1,3,2-dioxaphosphepane[10] in the presence of 9 mol. equiv. of tetrazole in CH_2Cl_2 solution at r.t. for 1.5 h followed by treatment with 1 mol. equiv. of MCPBA at -60°C for 10 min. and at r.t. for another 10 min. to yield the trisphosphate **5**, an oil, $\delta^{31}P(C_6D_6)$ -2.06, -1.55 and -0.84 ppm, 90%. Finally, hydrogenolysis of the benzyl groups over 10%-Pd/C in CH_3OH for 5h followed by acetal deprotection with AcOH-H_2O (1:1) at r.t.(18h) converted **5** into Ins(1,4,5)P_3, which was isolated and identified as a hexasodium salt.[11]

Scheme 2.



(i) Bu^tSiMe_2Cl, imidazole, acetonitrile; (ii) 2,2-dimethoxypropane, DMF, PPTS, TsOH; (iii) Bu_4NF/THF; (iv) a: 2-dimethylamino-5,6-benzo-1,3,2-dioxaphosphepane, tetrazole, CH_2Cl_2, b: MCPBA; (v) H_2/Pd/C; (vi) H_2O-AcOH 1:1.

As shown in Scheme 2, treatment of **1** with 2.2 mol. equiv. of *tert*-butylchlorodimethylsilane in the presence of 5 mol. equiv. of imidazole in acetonitrile solution at r.t. for 8h afforded diol **6**, an oil, $[\alpha]_D$=-10° (c 2.0, $CHCl_3$), in 50% yield. Diol **6** was then reacted with an excess of 2,2-dimethoxypropane in DMF in the presence of catalytic amounts of TsOH and PPTS (r.t.,2.5h), to give fully protected inositol derivative **7**, an oil, $[\alpha]_D$=+3° (c 2.0, $CHCl_3$), in 90% yield. Cleavage of silyl ethers by treatment of **7** with 1.1M Bu_4NF in THF at r.t. for 2h led to the formation of the bisacetal **8**, a glass, $[\alpha]_D$=+26.5° (c 2.2 $CHCl_3$), 100%, which was subsequently subjected to phosphorylation in the same way as described above for **4**. The resulting bisphosphate **9** [an oil, $\delta^{31}P(C_6D_6)$ -2.22 and -1.28 ppm, $[\alpha]_D$=+1.1° (c 2.2, $CHCl_3$), 99%], was finally converted into the title product in 80% yield by successive treatment with H_2 over 10%-Pd/C in CH_3OH at r.t. for 5h, and AcOH-H_2O (1:1) at r.t. for 20h. The Ins(1,4)P_2 thus obtained was isolated and characterized in the form of its tetracyclohexylammonium salt.[12]

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REFERENCES AND NOTES

1. For recent reviews see: Berridge, M.J.; Irvine, R.F. *Nature* **1989**, *341*, 197. Downes, C.P.; Macphee, C.H. *Eur. J. Biochem.* **1990**, *193*, 1.
2. Agranoff, B.W.; Bradley, R.M.; Brady, R.O. *J.Biol.Chem.* **1958**, *233*, 1077. Paulus, H.; Kennedy, E.P. *J.Biol.Chem.* **1960**, *235*, 1303.
3. For previous syntheses of D-*myo*-inositol 1,4,5-trisphosphate see: a) Ozaki, S.; Watanabe, Y.; Ogasawara, T.; Kondo, Y.; Shiotani, N.; Nishii, H.; Matsuki, T. *Tetrahedron Lett.* **1986**, *27*, 3157. b) Reese, C.B.; Ward, J.G. *Tetrahedron Lett.* **1987**, *28*, 2309. c) Vacca, J.P.; deSolms, S.J.; Huff, J.R. *J.Am.Chem.Soc. 1987*, *109*, 3478. See also ref. 4b. d) Liu, Y.C.; Chen, C.S. *Tetrahedron Lett.* **1989**, *30*, 1617. e) Stepanov, A.E.; Runova, O.B.; Schlewer, G.; Spiess, B.; Shvets, V.I. *Tetrahedron Lett.* **1989**, *30*, 5125. f) Ley, S.V. *Pure Appl. Chem.* **1990**, *62*, 2031.
4. Only two syntheses of D-*myo*-inositol 1,4-bisphosphate are on record a) Krylova, V.N.; Kobelkova, N.I.; Oleinik, G.F.; Shvets, V.I. *Zh.Org.Khim.* **1980**, *16*, 62., b) Vacca, J.P.; deSolms S.J.; Huff, J.R.; Billington, D.C.; Baker, R.; Kulagowski, J.J.; Mawer, I.M. *Tetrahedron* **1989**, *45*, 5702.
5. Bruzik, K.S.; Salamończyk G.M. *Carbohydr. Res.* **1989**, *195*, 67.
6. Salamończyk G.M.; Pietrusiewicz K.M. *Tetahedron Lett.* **1991**, *32*, 000.
7. In harmony with the recently suggested selectivity of bis-silylation of a closely related 2,3-*O*-cyclopentylidene-*myo*-inositol; Cf. ref. 3b.
8. With the exception of the final products which were isolated by precipitation of their salts, all compounds reported in this communication were isolated by column chromatography and were adequately analyzed by ^{1}H, ^{13}C and ^{31}P NMR techniques including DEPT and 2D COSY experiments when required. Full details of the exploratory experiments will be given in the forthcoming full account.
9. Watanabe, Y.; Komoda, Y.; Ebisuya, K.; Ozaki, S. *Tetrahedron Lett.* **1990**, *31*, 255.
10. This reagent can be conveniently prepared from commercial hexamethylphosphorous triamide by the published procedure: Arbuzov, B.A.; Kadyrov, R.A.; Klochkov, V.V.; Arshinova, R.P.; Aganov, A.V. *Izv. Akad. Nauk SSSR, Ser. Khim.* **1982**, 588.
11. **Ins(1,4,5)P_3**: NMR(D_2O pD 9.5) $\delta^{31}P$ 3.63, 5.31, 5.41 ppm; $\delta^{13}C$ 73.24, 74.34, 75.17, 77.21, 77.67, 79.86 ppm; $\delta^{1}H$ 3.71(dd, J=9.8, 3.0 Hz), 3.90(m, 3H), 4.15(q, J=9.0 Hz), 4.33(brt, J≅2.0 Hz) ppm; $[\alpha]_D$=-30°(c 0.8, H_2O pH 9.5), [Lit.[3c] $[\alpha]_D$=-30°(c 0.16, H_2O, pH 9.5)].
12. **Ins(1,4)P_2**: m.p. 178-182°C, [Lit.[4a] 174-176°C, Lit.[4b] 184-194°C(dec)]: NMR(D_2O) $\delta^{31}P$ 4.28, 4.92, ppm; $\delta^{13}C$ 27.21(8C), 27.71(4C), 33.76(8C), 53.72(4C), 74.47, 74.78, 75.57, 77.70(2C), 79.52 ppm; $\delta^{1}H$1.10-1.98-(m,40H), 3.02-3.18(m,4H), 3.41(t, J=10.0 Hz), 3.61(dd, J=3.0, 10Hz), 3.76(t, J=9.55 Hz), 3.87(dt, J=3.0, 10 Hz), 4.07(q, J=3.83 Hz), 4.17-(t, J=2.8 Hz) ppm; $[\alpha]_D$=+1.8°(c 5.0, H_2O), [Lit.[4a] $[\alpha]_D$=+3.1°(c 2.0, H_2O), Lit.[4b] $[\alpha]_D$=+0.12°(c 4.0, H_2O)].

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