*Tetrahedron Letters*, Vol. 35, No. 1, pp. 123-124, 1994
Printed in Great Britain

0040-4039/94 $6.00 + .00
Pergamon Press Ltd

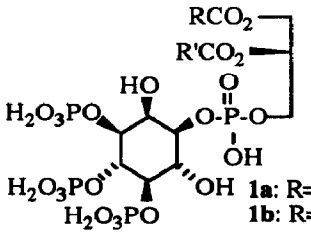
# Synthesis of a Phosphatidylinositol 3,4,5-Trisphosphate

**Yutaka Watanabe,* Hajimu Hirofuji, and Shoichiro Ozaki**

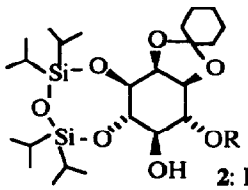
Department of Applied Chemistry, Faculty of Engineering, Ehime University, Matsuyama 790, Japan

**Abstract:** A phosphatidylinositol 3,4,5-trisphosphate was synthesized in a regioselective manner via 1,2-*O*-cyclohexylidene-3,4-*O*-(1,1,3,3-tetraisopropyldisiloxanyl)-*myo*-inositol readily derived from *myo*-inositol in two steps.

In addition to phosphatidylinositol 4,5-bisphosphate which is hydrolyzed to two intracellular second messengers, diacylglycerol and inositol 1,4,5-trisphosphate, 3-*O*-phosphorylated inositolphospholipids such as 1D-1-*O*-(3-*sn*-phosphatidyl)-*myo*-inositol 3,4,5-tris(phosphate) **1a** have more recently attracted considerable attention due to the expectation of their participation in cell proliferation and transformation.[1] These novel inositolphospholipids are generated from the corresponding 3-OH derivatives[2] by the action of phosphatidylinositol 3-kinase associated with the tyrosine kinase-linked receptor.[3] Falck and Abdali presented the chemical synthesis of **1b**, an analog of **1a** at the 200th National Meeting of the American Chemical Society in 1990.[4] In this communication, we report a concise synthesis of **1b**.



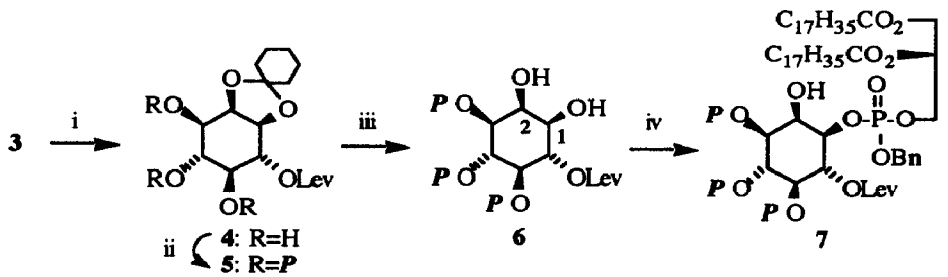
**1a**: R=stearoyl, R'=arachidonoyl
**1b**: R=R'=stearoyl



**2**: R=H
**3**: R=$CH_3CO(CH_2)_2CO$

For the present purpose, we chose 1,2-*O*-cyclohexylidene-3,4-*O*-(1,1,3,3-tetraisopropyldisiloxanyl)-*myo*-inositol **2** as a key synthetic intermediate which was derived regioselectively by the reaction of 1,2-cyclohexylidene-*myo*-inositol with the corresponding dichlorodisiloxane reagent.[5]

Diol **2** was selectively converted to 6-levulinyl (4-oxopentanoyl) ester **3** (levulinic acid, 1,3-dicyclohexylcarbodiimide, 4-dimethylaminopyridine; 84% yield) which was then desilylated to furnish triol **4**. The triol derivative was phosphorylated via phosphoramidite approach to give tris(dibenzyl phosphate) **5** in 72% yield. Careful removal of the cyclohexylidene group in **5** was achieved by treatment with trifluoroacetic acid and commercial dichloromethane containing a small amount of methanol as a stabilizer at 0 °C to give 1,2-diol **6** in 83% yield without affecting the other functional groups. Diol **6** was then phosphorylated regioselectively via the phosphonium salt methodology recently reported from this laboratory.[6] Thus, **6** was treated with dibenzyl 1,2-distearoyl-*sn*-glyceryl phosphite which was prepared by the 1*H*-tetrazole catalyzed reaction of the corresponding glycerol with dibenzyl *N,N*-diisopropylphosphoramidite in the presence of pyridinium bromide perbromide and



i. $n$-$Bu_4NF{\cdot}H_2O$, $PhCO_2H$ (85%); ii. $(PhCH_2O)_2PN^iPr_2$, 1*H*-tetrazole, then mCPBA (72%); iii. $CF_3CO_2H$, commercial $CH_2Cl_2$ (83%); iv. dibenzyl 1,2-distearoyl-*sn*-glyceryl phosphite, Py·HBr$_3$, 2,6-dimethylpyridine (36%). *Abbreviations*: Lev=$CH_3CO(CH_2)_2CO$; ***P*** =$(BnO)_2P(O)$; Bn=benzyl; Py=pyridine

2,6-dimethylpyridine to afford 1-phosphorylated product **7** in 36% yield.[7] The $^1H$ NMR of the phosphate gave broad peaks due to a mixture of four diastereomers. But the facts that **6** was converted regioselectively to 1-dibenzyl phosphorylated derivative[8] by the reaction with tribenzyl phosphite in a similar manner and that the chemical shift for $H_2$ in the $^1H$ NMR spectrum was similar to that for **7** supported the proposed phosphorylation site in **7**. The final deprotection was achieved by hydrogenolysis on 5% Pd-C in ethyl acetate at room temperature overnight (quantitative) followed by hydrazinolysis of the levulinyl group (77% yield). **1b** was obtained as crystalline solid after washing with acetone.[9]

We have now accomplished the much shorter synthesis of phosphatidylinositol 3,4,5-trisphosphate than Falck and Abdali's procedure although the inositol moiety in ours is racemic.

**Acknowledgment:** This work was partially supported by Tokuyama Science Foundation and a Grant-in-Aid for Scientific Research from the Ministry of Education, Science and Culture of Japan (No. 04640529).

## REFERENCES AND NOTES

1. For a review, see Berridge, M. J. *Nature*, **1993**, *361*, 315-325.
2. Whitman, M.; Downes, C. P.; Keeler, M.; Keller, T.; Cantley, L. *Nature*, **1988**, *332*, 644-646. Auger, K. R.; Serunian, L. A.; Soltoff, S. P.; Libby, P.; Cantley, L. C. *Cell*, **1989**, *57*, 167-175.
3. Talmage, D. A.; Freund, R.; Young, A. T.; Dahl, J.; Dawe, C. J.; Benjamin, T. L. *Cell*, **1989**, *59*, 55-65.
4. Falck J. R.; Abdali, A.: Enantiospecific Synthesis of Inositol Polyphosphates, Phosphatidylinositides, and Analogues from (-)-Quinic Acid. In *Inositol phosphates and Derivatives, Synthesis, Biochemistry, and Therapeutic Potential*; Reitz, A. B. Ed.; American Chemical Society; Washington, DC, 1991; pp. 145-154.
5. Watanabe, Y.; Mitani, M.; Morita, T.; Ozaki, S. *J. Chem. Soc., Chem. Commun.* **1989**, 482-483.
6. Watanabe, Y.; Inada, E.; Jinno, M.; Ozaki, S. Tetrahedron Lett. **1993**, *34*, 497-500.
7. FabMs m/z 1837.05 $[M+H]^+$: $\delta_H$ 0.85 (t, *J* 6.70 Hz, 6H), 0.98-1.37 (m, 56H), 1.53 (br, 4H), 2.18 (s, 1H), 2.18-2.37 (m, 4H), 2.37-2.68 (m, 4H), 3.90-4.50 (complex, 7H), 4.65 (br, 1H=$H_2$), 4.80-5.29 (complex, 16H), 5.60 (m, 1H), and 7.08-7.44 (m, 35H); $\delta_P$ -1.81 and -1.76 (3/4P), -1.69 (3/4P), -1.57 and -1.56 (0.5P), -1.26, -1.20, -1.18 and -1.15 (1P), and -0.68 (1P).
8. $\delta_P$ -1.75, -1.45, -1.11, and -0.64 ppm; $\delta_H$ (partial) 4.12 (dt, *J* 2.44 and 9.76 Hz, 1H=$H_1$), 4.68 ppm (t, *J* 2.44 Hz, 1H=$H_2$). These data suggest that **6** was phosphorylated at OH-1 and not at 2.
9. FabMs m/z 1105.4 $[M-H]^-$: $R_f$($CHCl_3$/$Me_2CO$/MeOH/AcOH/$H_2O$ 40:15:13:12:8) 0.3: mp ca. 140 °C: $\delta_H$ 0.95 (t, *J* 6.70 Hz, 6H), 1.08-1.42 (m, 56H), 1.60 (br, 4H), 2.22-2.40 (m, 4H), 3.62-3.74 (m, 2H), 4.00-4.43 (complex, 8H), and 5.28 (br, 1H); $\delta_P$ -0.96, 0.15, 0.61, and 0.76.

(*Received in Japan* 28 *April* 1993; *accepted* 13 *August* 1993)