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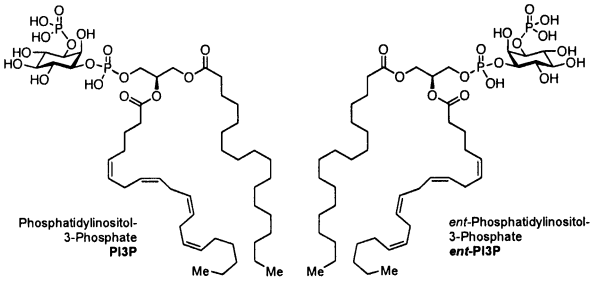
Asymmetric Syntheses of Phosphatidylinositol-3-Phosphates with Saturated and Unsaturated Side Chains through Catalytic Asymmetric Phosphorylation

Bianca R. Sculimbrene, Yingju Xu, and Scott J. Miller*

Department of Chemistry, Merkert Chemistry Center, Boston College, Chestnut Hill, Massachusetts 02467-3860

Received June 9, 2004; E-mail: scott.miller.1@bc.edu

The chemistry and biology of the phosphatidylinositol phosphates (PIPs) have emerged as a complex field due in part to the ubiquity of PIP-dependent cellular signaling.[1] In addition, the field has benefited from synthetic studies of these molecules, with natural products, synthetic probes, and analogues fueling chemical biological investigations.[2] Themes of the syntheses reported to date include complex, multistep schemes resulting from manipulations of chiral pool precursors such as D-glucose, the inositol ring system, which contains six stereochemically unique sites, and/or classical resolutions.[3,4] A significant opportunity for study of atomic level, high-resolution ligand−receptor interactions involving the PIPs and their targets would be enhanced if highly efficient syntheses of PIPs and their analogues, in each enantiomeric series, could be developed. We report herein initial steps along these lines, culminating in rapid total syntheses of optically pure **PI3P** and *ent*-**PI3P** with both saturated and unsaturated side chains. **PI3P** has been implicated as a product of phosphoinositide-3-kinase (PI3K), an important player in the biochemistry of cell cycle progression.[5] On the basis of catalytic asymmetric phosphorylation, the schemes reported herein provide access to either enantiomer of **PI3P** with equal efficiency, on the basis of the choice of synthetic catalyst.

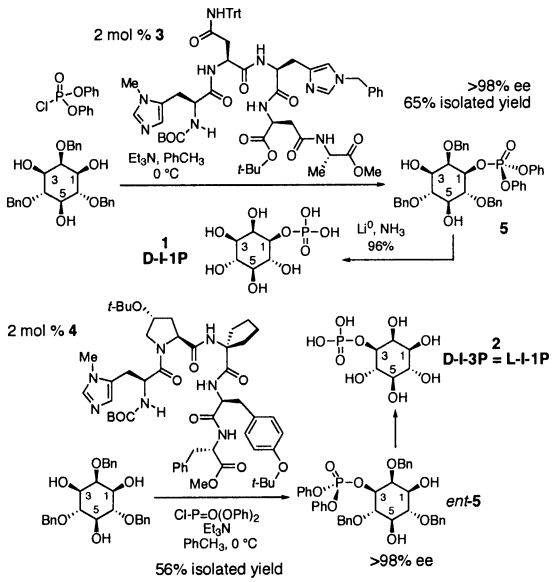


As a first step toward improved access, we recently reported an approach to the syntheses of D-*myo*-inositol-1-phosphate (D-I-1P, **1**), and its enantiomer D-*myo*-inositol-3-phosphate (D-I-3P = L-I-1P, **2**), employing peptide-catalyzed asymmetric phosphorylations as the key step.[6] For this purpose, we discovered peptides **3** and **4**, which exhibited enantiodivergent catalysis as exhibited in Scheme 1, affording desymmetrized phosphates **5** and *ent*-**5** with high optical purity. A critical component of our strategy was the decision that the synthetic schemes should originate with inositol, an achiral, *meso* precursor that is readily available.

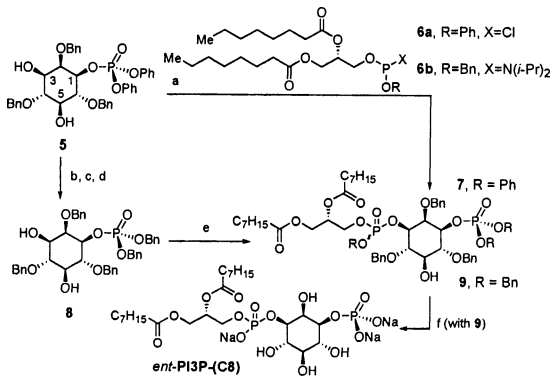
Our initial target for synthesis was *ent*-**PI3P-(C8)** with saturated C8-side chains on the glycerol unit. **PI3P-(C8)** has been a staple of chemical biological study. This compound also constitutes a more straightforward target strategically since the saturated side chains are compatible with global deprotection by hydrogenolysis.

Direct synthesis of the *ent*-**PI3P-(C8)** analogues from intermediate **5** required a swap of the phosphate ester protecting groups (Scheme 2). Initially, desymmetrized phosphate **5** was subjected to standard chlorophosphite coupling with **6a** to deliver fully

Scheme 1



Scheme 2 [a]



[a] Conditions: (a) **6a**, Hunig's Base, THF, −78 °C; then 30% H_2O_2/H_2O, 31%; (b) TBSCl, imidazole, DMF, 89%; (c) NaH, BnOH, THF, 84%; (d) HF pyridine, THF, 86%; (e) dicyanoimidazole, toluene/CH_2Cl_2, (1:1), **6b**; then 30% H_2O_2/H_2O; (f) H_2, $Pd(OH)_2$/C, *t*-BuOH/H_2O, $NaHCO_3$, 85%.

protected *ent*-**PI3P-(C8)** analogue **7** (31% isolated yield).[7,8] Compound **7** is, in principle, one step from a fully deprotected *ent*-**PI3P-(C8)**, provided that catalytic cleavage of both the benzyl ethers and phenyl phosphate esters could be achieved in one step. However, under the conditions we explored (e.g., H_2, PtO_2), benzyl ring hydrogenation intervened. As a result, a high-yielding three-step sequence was developed to convert phenyl phosphate **5** to benzyl phosphate **8**. Coupling to phosphoramidite **6b** then delivered **9**,[7,9] which was readily converted to *ent*-**PI3P-(C8)** in 85% yield. Of note, the identical sequence initiated with the desymmetrization of the *meso*-2,4,6-tris(*O*-benzyl)-*myo*-inositol substrate with peptide

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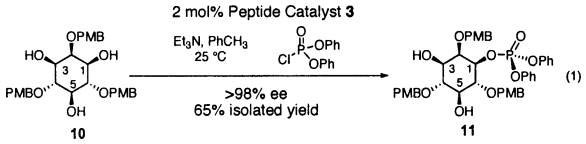
4 was carried out to provide the naturally occurring enantiomer of **PI3P-(C8)**. (See Supporting Information for details.) These schemes, despite the phosphate ester swap, may represent the most direct access to **PI3P-(C8)** targets reported to date.

We then turned our attention to the more ambitious objective of preparing **PI3P** compounds with the naturally implicated arachidonate ester side chains. For this objective, clearly the overall protecting group scheme would require alteration, as catalytic hydrogenation would surely reduce the alkenes resident in the side chains. After considerable experimentation, we found that *para*-methoxybenzyl (PMB) ethers were compatible with a similarly direct scheme.

The first question to answer was the viability of catalytic asymmetric phosphorylation of substrate **10**, with the tris(PMB)-array on the inositol ring. As before, peptide **3** proved to be highly effective in the desymmetrization reaction, producing phosphate **11** in 65% isolated yield, with >98% ee (eq 1). Of note, the catalytic phosphorylation proceeded at ambient temperature without a decrease in selectivity. We also found that the desymmetrization may be conducted with 1 mol % catalyst without loss of efficiency.

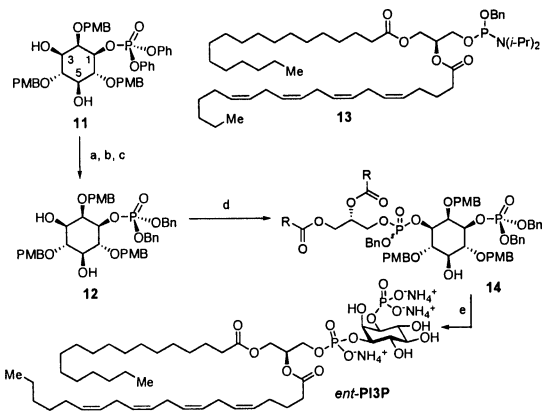


Phosphate **11** turned out to move through the phosphate ester swap with equal facility in comparison to phosphate **5**. Thus, bis-silylation followed by transesterifcation and desilylation afforded phosphate **12** (Scheme 3). Coupling of **12** to the arachidonic acid-derived phosphoramidite **13**[10] afforded protected *ent*-**PI3P** with the unsaturated side chains installed (**14**). As before, the yield of coupled product **14** is somewhat compromised by competitive phosphitylation at the 5-position of the inositol ring. Nevertheless, chromatographic purification delivers pure **14** as a mixture of phosphotriester diastereomers.

The deprotection of **14** to deliver *ent*-**PI3P** with the unsaturated side chains intact was achieved through careful optimization of reaction conditions. Ultimately, one-step treatment of **14** with 20 equiv of TMSBr in toluene at 70 °C for 12 h delivered *ent*-**PI3P**.[11] Initial attempts to achieve the deprotection in two steps (e.g., DDQ oxidation of the PMB groups followed by TMSBr-mediated cleavage of the benzyl phosphate esters) resulted in complicated reaction mixtures, perhaps related to phosphate migration. In addition, isolation of the final product proved to be difficult coming out of the DDQ-promoted oxidation. Instead, the one-step protocol enabled efficient removal of both the PMB and benzyl groups, delivering a product that was amenable to direct analysis by mass spectrometry and ^{1}H and ^{31}P NMR (solvent: $CD_3OD/CDCl_3/D_2O$, 4:3:1). In addition, in accord with the original characterization of these molecules, we found that we were able to purify these amphiphilic compounds by conventional chromatography.[12]

In summary, we report enantioselective total syntheses of **PI3P-(C8)** in each enantiomeric series, in addition to a synthesis of *ent*-**PI3P** with the arachidonate side chain in place. The sequences are rapid in terms of their overall step count and ease of operation. Furthermore, because the syntheses depend on asymmetric catalysis, the routes may be conducted in either enantiomeric series with comparable facility and economic considerations. These syntheses may provide an opportunity to deliver improved access to optically

Scheme 3 [a]



[a] Conditions: (a) TBSCl, imidazole, DMF, 89%; (b) NaH, BnOH, THF 99%; (c) HF pyridine, THF, 77%; (d) dicyanoimidazole, toluene/CH_2Cl_2 (1:1), **13**; then 30% H_2O_2/H_2O, 39%; (e) TMSBr (20 equiv)/$PhCH_3$, 70 °C; then NH_4OH, 61%.

pure targets and analogues in this family of natural products. In addition, they serve as a starting place for the development of other asymmetric phosphorylation catalysts that may provide direct access to alternative isomers and more highly phosphorylated targets in this series. Finally, these routes may prove to be useful in the preparation of **PI3P** analogues of interest for chemical biological study.

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Supporting Information Available: Experimental procedures and product characterization for all new compounds synthesized (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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