Introduction

Cidofovir (HPMPC) is an injectable antiviral medication used as a treatment for cytomegalovirus (CMV) retinitis (an infection of the retina of the eye) in people with AIDS and has now found application in the treatment of other DNA viruses. The drug first received FDA approval in 1996.

The active metabolite, cidofovir diphosphate, inhibits viral replication by selectively inhibiting viral DNA polymerases. It also inhibits human polymerases but this action is 8-600 times weaker than its actions on viral DNA polymerases. It also incorporates itself into viral DNA hence inhibiting viral DNA synthesis during reproduction.

It possesses in vitro activity against the following viruses:

- Human herpesviruses
- Adenoviruses
- Human poxviruses (including the smallpox virus)
- Human papillomavirus


The procedure used is an adaptation of the most concise preparation: Vemishetti et al., Tetrahedron Letters, 35, 1994, 3243-46, which summarizes the original patent: PCT/US91/05578, 20 February, 1992.
PART 1: Preparation of triphenylmethoxymethyloxirane

A 100 mL round bottom flask was charged with trityl chloride (2.66 g, 9.6 mmol) and CH$_2$Cl$_2$ (10 mL). It was cooled to ~0 °C under N$_2$ and then treated with triethylamine (1.42 g, 14 mmol). After stirring for 1 hour at ~0 °C, a solution of glycidol (0.74 g, 10 mmol) in CH$_2$Cl$_2$ (2.5 mL) was added over 0.75 hour. The resulting solution was allowed to warm to ambient temperature and stirred for 3 hours. The mixture was then filtered, and the filtrate washed with water (2 x 25 mL) and brine (1 x 25 mL). The organic phase was then dried over MgSO$_4$ and concentrated to a foam, which on crystallization from isopropyl alcohol gave the title compound as an off-white power.

A $^1$H NMR spectrum and melting point are required at this point before you proceed to the next step.

PART 2: Preparation of N$^4$-benzoyl-N$^1$-[(2-hydroxy-3-triphenylmethoxy)-propyl]cytosine

To N$^4$-benzoylcytosine (1.00 g, 4.7 mmol) in dry DMF (10 mL) at 100 °C under N$_2$ was added NaH (dispersion in mineral oil, 1.2 mmol) in one portion and the slurry stirred for 0.25 hour. Triphenylmethoxymethyloxirane (1.25 g, 4.0 mmol) was added and further stirred at 100 °C for 4 hours. The reaction mixture is filtered and the filtrate used in the next step without purification.

To confirm that the desired product is present in sufficient purity, an LC-MS will be performed.
PART 3: Preparation of N⁴-benzoyl-N¹-[[(diethylphosphonylmethoxy)-3-triphenylmethoxy]-propyl]cytosine

The crude DMF solution from the previous procedure was cooled to 0 °C under N₂ in a 50 mL round bottom flask. NaH (dispersion in mineral oil, 11 mmol) was added in two portions. Immediately thereafter was added diethyl tosylxymethylphosphonate (1.74 g, 5.4 mmol) and mixture was stirred for 6 hours. The reaction mixture was diluted with ethyl acetate (25 mL) and washed with water (2 x 12 mL) and saturated NaHCO₃ (12 mL), dried over MgSO₄ and concentrated to afford the crude title compound.

A ¹H NMR spectrum is required at this point before you proceed to the next step. DMSO-d₆ will be the solvent.

PART 4: Preparation of N⁴-benzoyl-N¹-[[(diethylphosphonylmethoxy)-3-hydroxy]-propyl]-cytosine

Generated HCl gas was bubbled into a solution of the crude product from Part 3 in CH₂Cl₂ (20 mL) at 0-5 °C for 15 minutes. Complete consumption of the starting material will be determined by LC-MS. Water (10 mL) was added and the two-phase mixture stirred vigorously for 10 minutes. The organic phase was separated and extracted with 10% HCl solution (2 x 5 mL). The combined aqueous solution was cooled to DMF solution from the last procedure was cooled to 0-5 °C and adjusted to pH = 8 with 40% NaOH solution and then extracted with CH₂Cl₂ (2 x 10 mL). The combined CH₂Cl₂ solution was dried over MgSO₄ and concentrated to give the title compound as a viscous oil.

A ¹H NMR spectrum is required at this point before you proceed to the next step. DMSO-d₆ will be the solvent.
PART 5: Preparation of N\textsuperscript{4}-benzoyl-N\textsuperscript{1}-[(diethylphosphonylmethoxy-3-triphenylmethoxy)-propyl]cytosine

A solution of N\textsuperscript{4}-benzoyl-N\textsuperscript{1}-[(diethylphosphonylmethoxy-3-hydroxy)-propyl]cytosine (1.88 g, 4.28 mmol) in CH\textsubscript{2}Cl\textsubscript{2} (12 mL) under N\textsubscript{2} was treated with bromotrimethylsilane (2.00 mL, 15.2 mmol) and the resulting mixture stirred for 18 hours. The mixture is concentrated to a residue which is redissolved in CH\textsubscript{2}Cl\textsubscript{2} and re-concentrated to the persilylated compound as a tan foam. The material is used in the next procedure to with no further purification.

A \textsuperscript{1}H NMR spectrum is required at this point before you proceed to the next step. DMSO-\textit{d}\textsubscript{6} will be the solvent.

N\textsuperscript{4}-benzoyl-N\textsuperscript{1}-[(diethylphosphonylmethoxy-3-hydroxy)-propyl]cytosine

PART 6: Preparation of N\textsuperscript{4}-benzoyl-N\textsuperscript{1}-[(diethylphosphonylmethoxy-3-hydroxy)-propyl]cytosine

The material from the previous procedure is dissolved in concentrated NH\textsubscript{4}OH (aq. NH\textsubscript{3}, aq., 10 mL) and stirred at room temperature for 4 hours. The aqueous reaction mixture was extracted with CH\textsubscript{2}Cl\textsubscript{2} (2 x 10 mL) to remove most of the benzamide (protecting group). The aqueous phase was then filtered and concentrated in vacuo until the solution was neutral. The concentrated solution was diluted with water to a volume of 8 mL and ethanol (6 mL) was added. Adjusting the pH to 3.0 with careful addition of HCl precipitated the product. The resulting thick slurry was stirred at room temperature for one hour and then stored at 0-5 °C for 16 hours. The solid product was collected by filtration, washed with aqueous ethanol (2:1, H\textsubscript{2}O:EtOH, 2 x 15 mL) and dried to a constant weight in vacuo to give the final anti-viral compound HPMPC.

Full characterization is required at this point, including \textsuperscript{1}H, \textsuperscript{13}C and \textsuperscript{31}P NMR (D\textsubscript{2}O solvent), DQFCOSY, DEPT-135, HSQC and HMBC, FT-ATR and UV-Vis. A melting point should also be determined.