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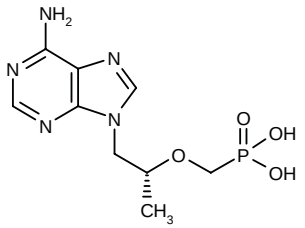
(*R*)-PMPA and Bis(POC)PMPA

Anti-HIV

(R)-PMPA

(*R*)-9-[2-(Phosphonomethoxy)propyl]adenine

(*R*)-[[2-(6-Amino-9*H*-purin-9-yl)-1-methylethoxy]methyl]-phosphonic acid



$C_9H_{14}N_5O_4P$ Mol wt: 287.2146

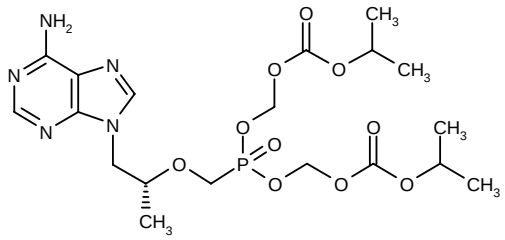
CAS: 147127-20-6
CAS: 147127-19-3 [as (*S*)-isomer]

EN: 224671

Bis(POC)PMPA

(*R*)-[[2-(6-Amino-9*H*-purin-9-yl)-1-methylethoxy]-methyl]phosphonic acid bis(isopropoxycarbonyloxy-methyl) ester

2-(Adenin-9-yl)-1(*R*)-methylethoxymethylphosphonic acid bis(isopropoxycarbonyloxymethyl) ester



$C_{19}H_{30}N_5O_{10}P$ Mol wt: 519.4450

CAS: 201341-05-1
CAS: 202138-50-9 [as fumarate salt (1:1)]

EN: 246665

Synthesis of (*R*)-PMPA

(*R*)-PMPA has been obtained by three related ways:

1) The protection of isobutyl D-(+)-lactate (I) with dihydropyran (DHP)/HCl in DMF gives the tetrahydropyranyloxy derivative (II), which is reduced with bis(2-methoxyethoxy)aluminum hydride in refluxing ether/toluene yielding 2(*R*)-(tetrahydropyranyloxy)-1-propanol (III). The tosylation of (III) with tosyl chloride as usual affords the expected tosylate (VI), which is condensed with adenine (V) by means of Cs_2CO_3 in hot DMF, affording 9-[2(*R*)-(tetrahydropyranyloxy)propyl]adenine (VI). The deprotection of (VI) with sulfuric acid affords 9-[2(*R*)-hydroxypropyl]adenine (VII), which is *N*-benzoylated with benzoyl chloride/chlorotrimethylsilane in pyridine to give the benzamide (VIII), which is condensed with tosyloxymethylphosphonic acid diisopropyl ester (IX) by means of NaH in DMF to yield 9-[2(*R*)-(diisopropoxyphosphorylmethoxy)propyl]adenine (X). Finally, this compound is hydrolyzed by means of bromotrimethylsilane in acetonotrile (1, 2). Scheme 1.

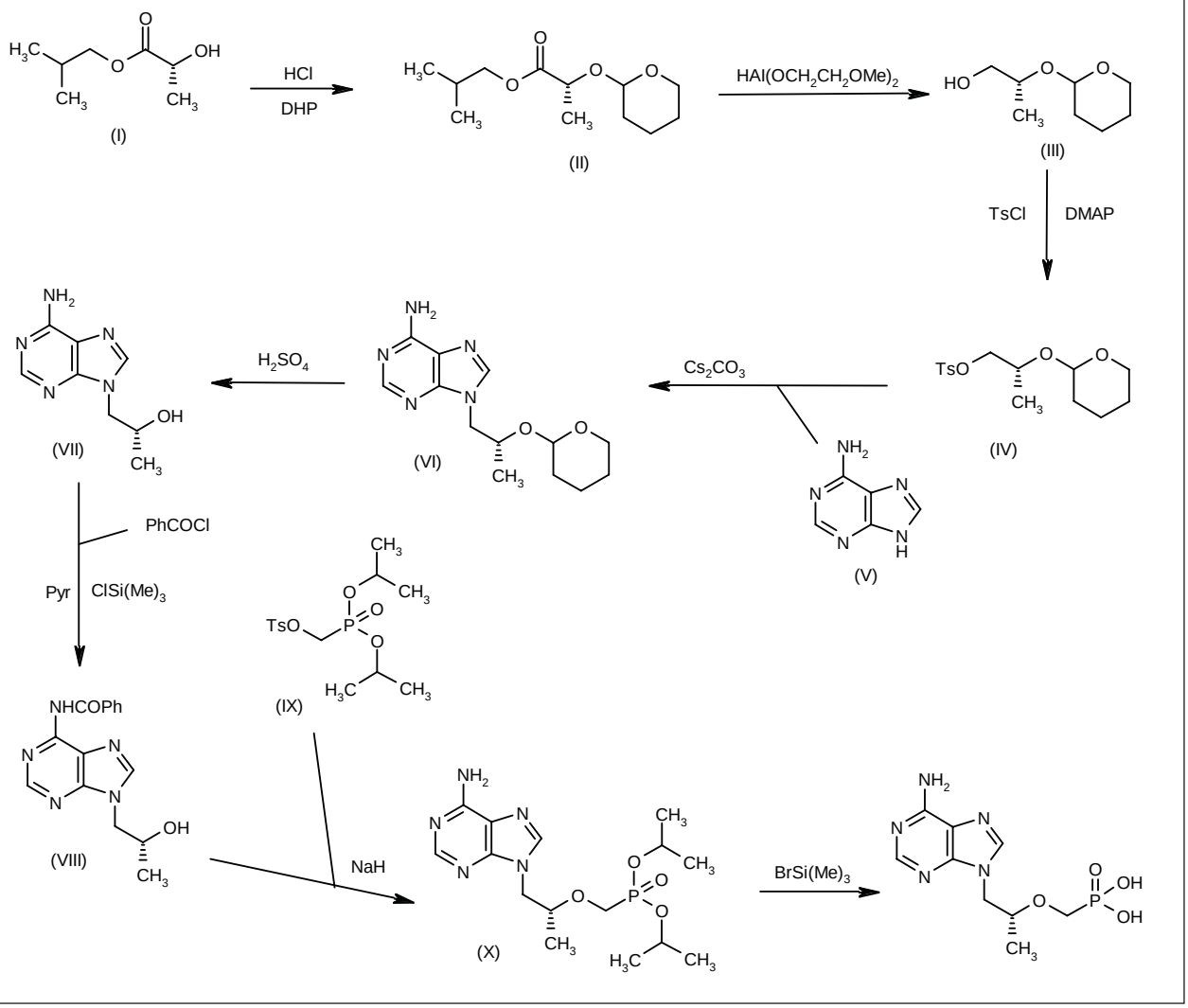
2) The reaction of the previously described (*R*)-2-(2-tetrahydropyranyloxy)-1-propanol (III) with benzyl bromide (XI) by means of NaH in DMF, followed by a treatment with Dowex 50X, gives 1-benzyloxy-2(*R*)-propanol (XII), which is condensed with tosyloxymethylphosphonic acid diisopropyl ester (IX) by means of NaH in THF, yielding 2-benzyloxy-1(*R*)-methylethoxymethylphosphonic acid diisopropyl ester (XIII). The hydrogenolysis of (XIII) over Pd/C in methanol affords 2-hydroxy-1(*R*)-methylethoxymethylphosphonic acid diisopropyl ester (XIV), which is tosylated with tosyl chloride/dimethylaminopyridine in pyridine to give the expected tosylate (XV). The condensation of (XV) with adenine (VI) by means of Cs_2CO_3 in hot DMF yields 9-[2(*R*)-(diisopropoxyphosphorylmethoxy)propyl]adenine (X), which is finally hydrolized as before (3). Scheme 2.

3) The catalytic hydrogenation of (*S*)-glycidol (XVI) over Pd/C gives the (*R*)-1,2-propanediol (XVII), which is esterified with diethyl carbonate (XVIII)/NaOEt, yielding the cyclic carbonate (XIX). The reaction of (XIX) with adenine (V) by means of NaOH in DMF affords 9-[2(*R*)-hydroxypropyl]adenine (VII), which is condensed with tosyloxymethylphosphonic acid diethyl ester (XX) by means of lithium *tert*-butoxide in THF, giving 9-[2(*R*)-(diethoxyphosphorylmethoxy)propyl]adenine (XXI). Finally, this compound is hydrolized with bromotrimethylsilane as before (4, 5). Scheme 3.

Compound (XX) is obtained by reaction of diethyl phosphite (XXII) with paraformaldehyde, yielding hydroxymethylphosphonic acid diethyl ester (XXIII), which is finally tosylated as usual (4). Scheme 3.

L.A. Sorbera, J. Castañer. Prous Science, P.O. Box 540, 08080 Barcelona, Spain.

Scheme 1: Synthesis of (*R*)-PMPA



Synthesis of Bis(POC)PMPA

The reaction of chloromethyl chloroformate (I) with isopropyl alcohol (II) by means of pyridine or triethylamine in ether gives the mixed carbonate (III), which is then condensed with (*R*)-PMPA (IV) by means of diisopropyl ethylamine in DMF (4, 6). Scheme 4.

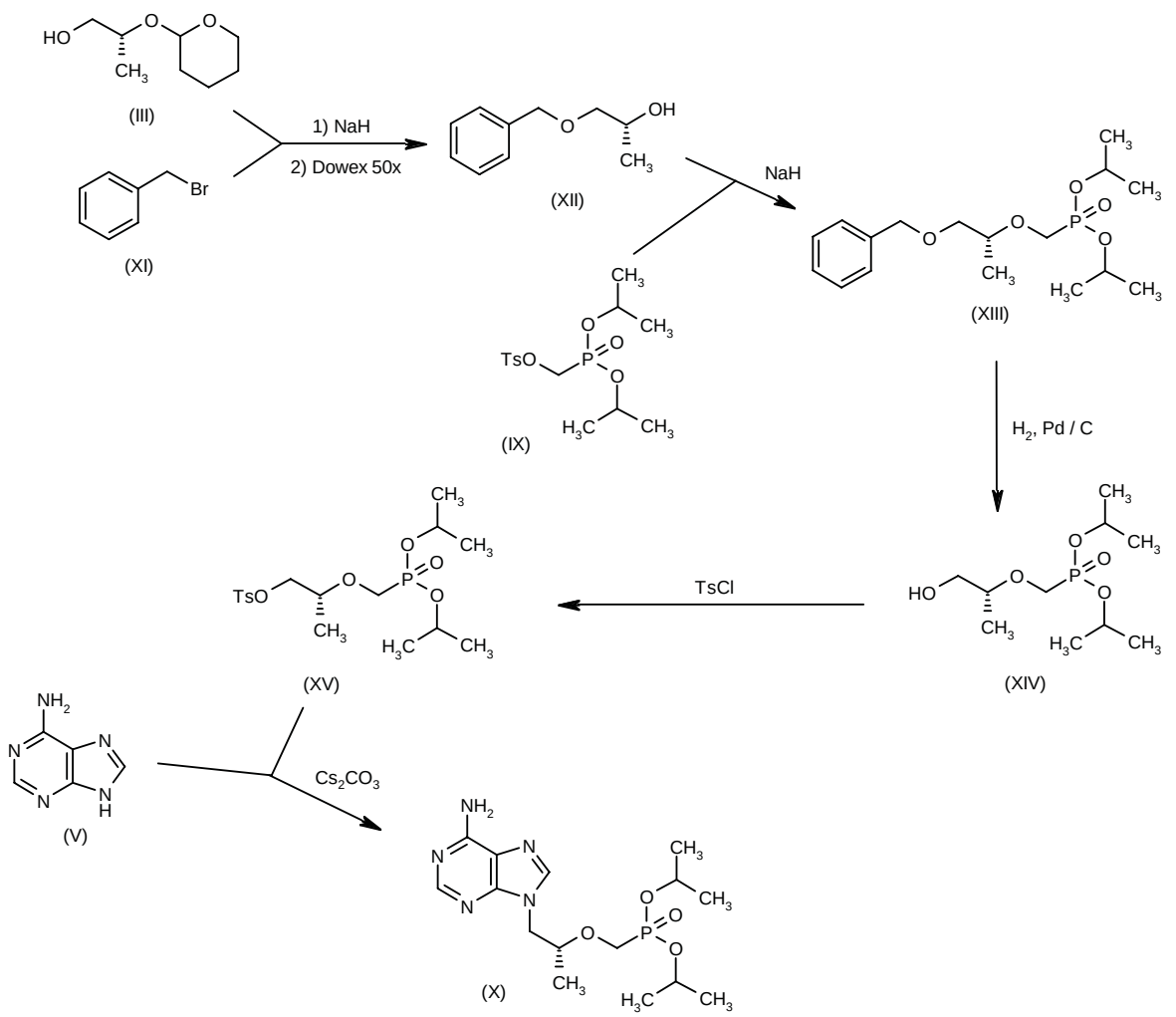
Introduction

A new class of compounds with significant antiviral activity were discovered during the course of structure-activity studies with acyclic phosphonate analogs of nucleotides. These compounds are *N*-(2-phosphonomethoxypropyl) derivatives of the heterocyclic base adenine (PMPA) and possess a phosphonomethylether functional group (1, 2, 7). The active enantiomer (*R*)-PMPA, distinguished from these studies, was found to have potent and selective activity against HIV and other retroviruses (1). In view of this agent's poor oral bioavailability in animals, several prodrugs of (*R*)-PMPA were prepared. One such prodrug, bis(POC)PMPA, a lipophilic ester derivative in an oral formulation, was chosen for further evaluation (6).

Pharmacological Actions

The *in vitro* antiviral activity of (*R*)-enantiomer of PMPA was found to be approximately 10-100 fold more potent than the (*S*)-enantiomer against HIV-1 (strains IIIb, Ba-L or HE) or HIV-2 (ROD) in Molt 4/clone 8 cells, human lymphocytes (MT-4), human monocytes,

Scheme 2: Synthesis of Intermediate (X)



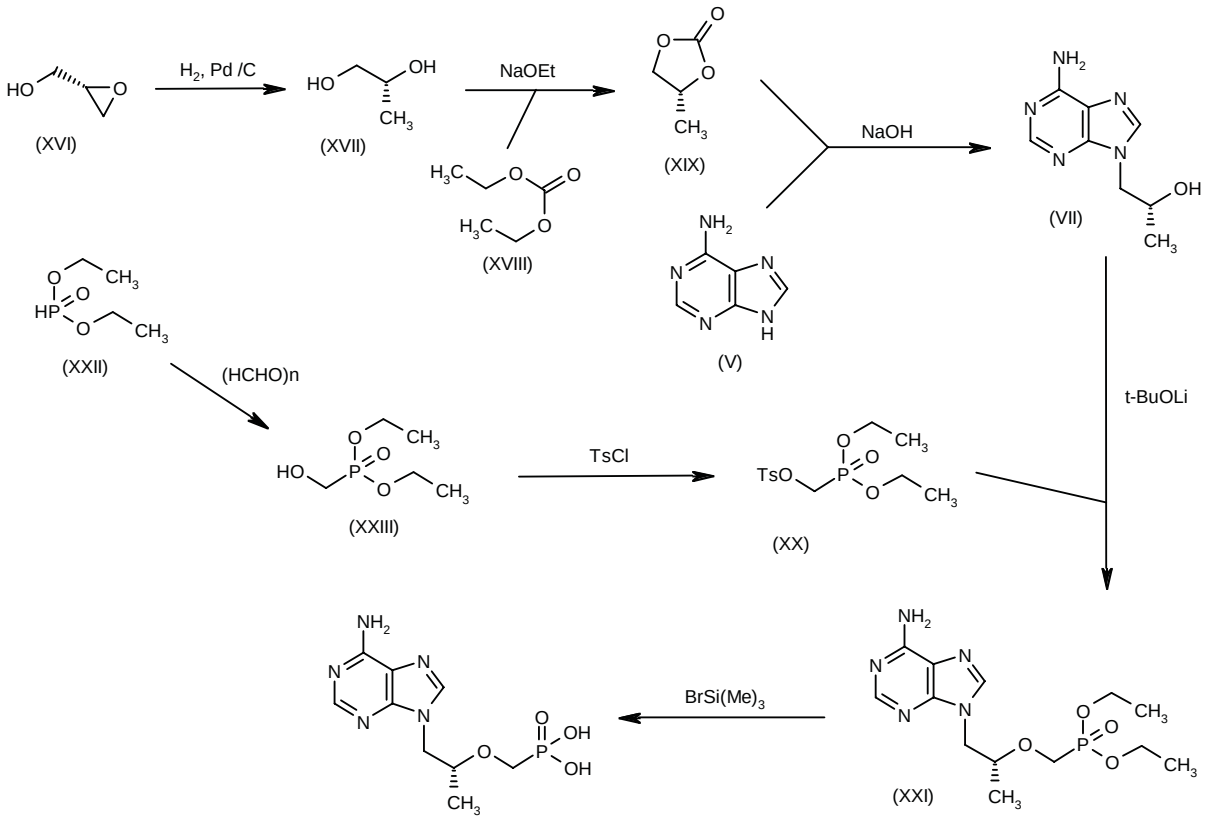
macrophages and peripheral blood mononuclear cells (PBMCs) (8). (*R*)-PMPA also displayed more selective antiviral effects in cell cultures infected with HIV-1 and HIV-2, feline immunodeficiency virus and Moloney murine sarcoma virus than MDL-74968 and 9-(2-phosphonylmethoxyethyl) derivatives of adenine (PMEA) and guanine (PMEG) (9). Moreover, when the antiviral activity against HIV-1/IIIb of (*R*)-PMPA and bis(POC)PMPA were compared *in vitro* in an infected T-lymphocytic MT-2 cell line, results showed that bis(POC)PMPA exhibited increased cytotoxicity with an IC_{50} of 0.0003 μM and a CC_{50} of 50 μM as compared to 0.5 and 250 μM, respectively, for (*R*)-PMPA. In addition to an enhanced selectivity index, bis(POC)PMPA also displayed improved chemical and enzymatic stability as compared to (*R*)-PMPA (6).

The improved efficacy of bis(POC)PMPA as compared to (*R*)-PMPA was further demonstrated in a recent *in vitro* study using MT-2 cells and PBMCs infected with HIV-1/IIIb; antiviral EC_{50}s of 0.63 and 0.18 μM for (*R*)-PMPA and 0.007 and 0.005 μM for bis(POC)PMPA were found in infected MT-2 cells and PBMCs, respectively. The greater than 100-fold antiviral activity exhibited by bis(POC)PMPA was shown to be due to rapid intracellular uptake producing a more than 1000-fold increase in intracellular accumulation of the active metabolite, PMPA diphosphate (PMPApp) (10).

Investigation of the antiviral activities of (*R*)-PMPA and bis(POC)PMPA against several drug-resistant HIV strains was performed *in vitro* using PBMCs and MT-2 cells. Results indicated that only the K65R strain was resistant to (*R*)-PMPA. K65R exhibited a reduced susceptibility to bis(POC)PMPA with inhibition still occurring at submicromolar, nontoxic concentrations of the prodrug (11).

In addition to potently inhibiting retroviruses, (*R*)-PMPA was found to have other effects *in vitro*. A study has described the efficacy of the agent against human

Scheme 3: Synthesis of (*R*)-PMPA



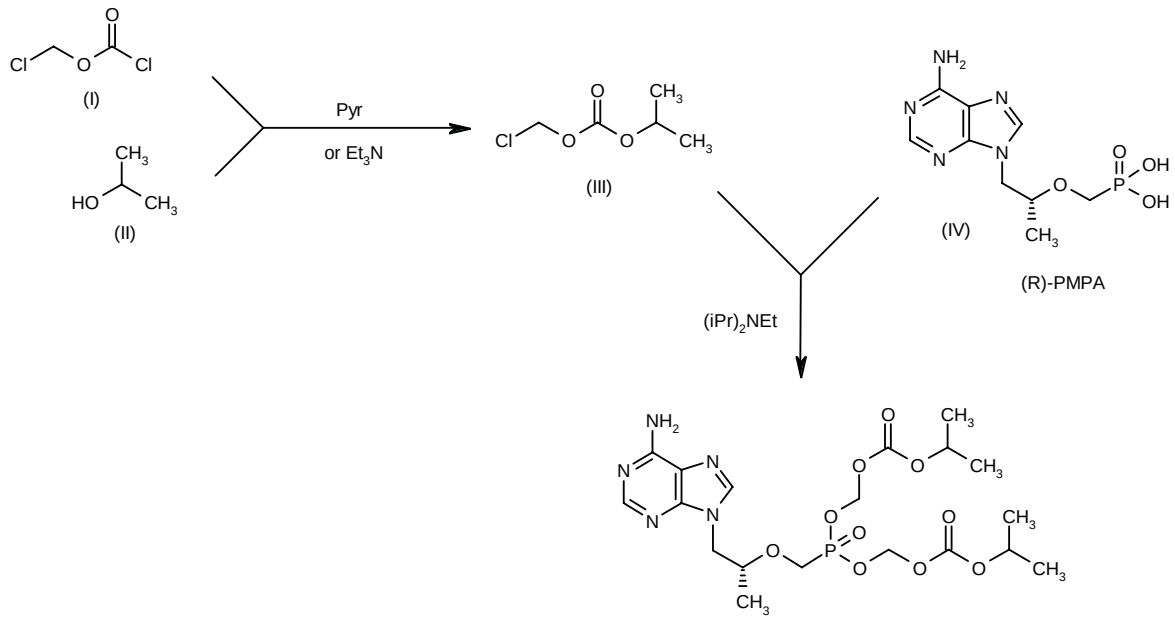
cytomegalovirus (HCMV) activity. (*R*)-PMPA was compared with other anti-CMV agents, cidofovir diphosphate and ganciclovir triphosphate, and the active diphosphate metabolite PMPApp was found to be a competitive inhibitor of dATP against HCMV DNA polymerase using activated calf thymus as a primer template (K_i = 0.62 ± 0.11 μM *vs*. 6.6 ± 0.8 μM for cidofovir). The V_{max}/K_m values for PMPApp against HCMV DNA polymerase when synthetic DNA oligomers were used as primer templates was 0.0092 pmol.min/unit/μM as compared to 0.014 and 0.0094 pmol.min/unit/μM for cidofovir diphosphate and ganciclovir triphosphate, respectively. Moreover, after 48 h of exposure of normal human epidermal keratinocytes to 5 μM (*R*)-PMPA, cells converted 59.5% of (*R*)-PMPA to PMPApp, demonstrating successful intracellular phosphorylation of the agent. These results suggest that PMPA may be a candidate for clinical trials investigating possible therapies for HCMV infections (12).

(*R*)-PMPA also displays immunomodulatory effects influencing the secretion of cytokines and production of nitric oxide (NO) in murine peritoneal macrophages *in vitro*. (*R*)-PMPA significantly stimulated secretion of IL-10 and TNF-α in a dose- and time-dependent manner with no effects on IFN-γ and IL-2 expression. In addition, IFN-γ-induced NO production was enhanced and studies using specific antibodies against TNF-α and IL-10 have shown that these cytokines mediate the (*R*)-PMPA-induced increase in NO production (13, 14).

The potent antiretroviral properties of (*R*)-PMPA shown *in vitro* have been confirmed *in vivo*. Five weeks following infection with the LP-BM5 murine leukemia virus known to progress to severe murine immunodeficiency (MAIDS), mice were administered (*R*)-PMPA (5 or 25 mg/kg i.p.). Progression to disease was prevented so that at 9 weeks none of the mice treated with 25 mg/kg (*R*)-PMPA developed severe MAIDS; only 1 animal was determined to have mild MAIDS (15).

A study has demonstrated that (*R*)-PMPA has potent inhibitory activity *in vivo* against acute and established feline immunodeficiency (FIV) with less toxicity than AZT or PMEA. Cats infected with FIV-B-2542 were treated with (*R*)-PMPA (30 mg/kg/day s.c. or 60 mg/kg /day p.o.) at the time of infection or 8 weeks after infection ensuring established FIV. FIV did not develop in any cats injected with (*R*)-PMPA at the time of infection and no hematologic or other toxicities were observed. In contrast, 100% of the placebo-treated cats developed FIV. Oral administration of (*R*)-PMPA was less effective, although a 40% pro-

Scheme 4: Synthesis of Bis(POC)PMPA



tection against progression of disease was afforded at the time of infection. In cats with established infections, (*R*)-PMPA treatment decreased plasma viral RNA levels although no significant effect was observed in viral levels from PBMC co-cultures from these animals (16).

Several preclinical studies have described the efficacy of (*R*)-PMPA *in vivo* against simian immunodeficiency virus (SIV) in adult and newborn macaques. Within less than 2 weeks of (*R*)-PMPA treatment (30 or 75 mg/kg/day s.c. for 28 days), SIV plasma and PBMC levels were reduced by more than 99% in adult chronically (for at least 19 weeks) SIV-infected cynomolgus macaques. (*R*)-PMPA at a dose of 30 mg/kg was well tolerated and increased $CD4^+$ counts without any observed adverse effects (17, 18). (*R*)-PMPA (20 or 30 mg/kg s.c. for 4 weeks) was also effective when administered 48 h prior to infection and 4 or 24 h after infection in adult macaques. SIV infection was prevented throughout the 56-week experimental period without adverse effects (19).

Preclinical studies have demonstrated that inoculation of newborn SIV-infected rhesus macaques with (*R*)-PMPA reduced the incidence of perinatal SIV infection. Newborns infected within 3 days after birth with oral SIVmac251 and/or SHIV-SF33 (i.v.) developed fatal immunodeficiency within 7 months of age, regardless of whether the pregnant mother had been administered a single injection of (*R*)-PMPA (30 mg/kg s.c.) 2 h prior to cesarean section. However, although 3/4 newborns infected within 2 h of delivery and immediately administrated (*R*)-PMPA (30/kg/day s.c.) for 2 weeks displayed initial transient disease, animals were healthy and seronegative by 8 months of age (20).

The long-term therapeutic and toxic effects of (*R*)-PMPA were also evaluated in SIV-infected newborn rhesus macaques. Reduction of viremia was observed in 4 macaques administered (*R*)-PMPA (30 mg/kg/day s.c.) beginning 3 weeks after infection; disease-free states were observed for more than 13 months and no adverse effects were noted. However, a virus with a 5-fold attenuated susceptibility to (*R*)-PMPA and associated with a K65R mutation emerged in all treated animals (21).

Pharmacokinetics and Metabolism

The pharmacokinetics of (*R*)-PMPA and the orally formulated prodrug, bis(POC)PMPA, have been described in several studies. Results using the human intestinal mucosal Caco-2 cell monolayer model show that the total transport of bis(POC)PMPA was 2.7% as compared to less than 0.1% for (*R*)-PMPA. The majority of metabolism of bis(POC)PMPA occurred within the epithelial cells with a relative resistance to degradation observed at the luminal side of the cells (22).

In PBMCs, bis(POC)PMPA displayed enhanced membrane permeability and rapid intracellular anabolism to PMPApp; accumulation of the active metabolite was observed in both resting and activated PBMCs. The half-life for accumulated intracellular PMPApp for both agents was 12-15 h and 33-50 h in activated and resting lym-

phocytes, respectively (10, 23). After incubation of PBMCs with 1 μM [^{3}H]bis(POC)PMPA for 1 h, the concentrations of PMPA, PMPApp and PMPA monophosphate (PMPAp) were 28, 1.9 and 6.3 μM, respectively (24).

The pharmacokinetics of both agents have been examined in mice and dogs. In one study, dogs were pretreated with pentagastrin and administered (*R*)-PMPA (10 mg/kg i.v. or p.o. or 1 mg/kg i.v. and 10 mg/kg i.p.); to determine metabolism and excretion of (*R*)-PMPA, 1 dog was injected with [^{14}C]-PMPA (10 mg/kg i.v.). The concentrations of plasma PMPA after i.v. injection had a terminal half-life of 10 h, and 70% of unchanged PMPA was excreted in urine; recovery from feces and bile was negligible. Active tubular secretion of PMPA was suggested to occur since the plasma clearance rate of the agent (0.28 ± 0.05 l/h/kg) was faster than the glomerular filtration rate for dogs. The bioavailability following oral and i.p. administration of 10 mg/kg was determined in this particular study to be approximately 17 and 74%, respectively (25). Other studies have reported that the oral bioavailability of (*R*)-PMPA in mice and dogs was < 3% (24). On the other hand, the oral bioavailability of bis(POC)PMPA in mice was determined to be 20-30% with efficient release of the active PMPA indicated, since neither the prodrug nor its monoester metabolite, mono(POC)PMPA, could be recovered in plasma after oral administration (22). Similarly, a bioavailability of 25-30% was found in dogs administered 60 mg/kg/day bis(POC)PMPA with marginal toxicity observed when administered for 5 days (24, 26). Further pharmacokinetic studies reported that bis(POC)PMPA had good chemical and intestinal stability when examined *in vitro* in dog intestinal, plasma and liver homgenates (27).

Clinical Studies

A randomized, placebo-controlled phase I/II study to evaluate the tolerability and efficacy of intravenously administered (*R*)-PMPA has shown that HIV RNA levels decreased continuously during dosing with final reductions of 0.6 and 1.1 log observed in HIV-infected patients receiving doses of 1 and 3 mg/kg, respectively; the reductions observed in the group receiving the higher dose were sustained for up to 1 week after treatment (28). Other phase I/II studies are in progress to assess the intravenous formulation of (*R*)-PMPA as a potential therapy for prevention of maternal/fetal HIV transmission and phase I trials are planned for evaluation of a topical formulation of (*R*)-PMPA (29).

Preliminary results have been reported from a phase I/II randomized, double-blind, placebo-controlled, dose-escalation study evaluating the safety and efficacy of oral bis(POC)PMPA monotherapy. Thirty-six fasted HIV-infected adults were administered a single dose of the prodrug (75, 150 or 300 mg/day p.o.) followed by a 7-day washout period and a subsequent 28-day dosing period. Bioavailability was found to increase with food intake from approximately 27% to 41%. The estimated serum half-life was equal to or greater than 17 h and systemic exposure was dose-dependent. After 28 days of treatment, dose-dependent reductions in HIV RNA levels from baseline were observed, with median log10 decreases of 0.32, 0.44, 1.22 and 0.06 for patients receiving 75, 150, 300 mg of bis(POC)PMPA and the placebo, respectively. Serious adverse effects included reversible elevations of creatine kinase in 18% and 14% of the prodrug- and placebo-treated patients, respectively, and 2 individuals experienced exacerbation of preexisting sensory neuropathies (30).

A phase I/II study is currently under way to evaluate whether the antiviral activity of oral bis(POC)PMPA therapy is enhanced in HIV-infected patients when administered in combination with hydroxyurea (31).

In addition, a phase II randomized, double-blind, crossover, dose- escalation study has been initiated to determine the efficacy of oral bis(POC)PMPA therapy in combination with other antiretroviral regimens. HIV-infected patients (n = 175) receiving a stable antiretroviral therapy of not more than three antiretroviral agents will be administered either a placebo or one of three oral doses of the prodrug for 48 weeks. After 24 weeks, those patients receiving the placebo will be eligible to cross over to open-label oral bis(POC)PMPA therapy (31).

Manufacturer

Gilead Sciences, Inc. (US).

References

1. Holy, A., Masojidkova, M. *Synthesis of enantiomeric N-(2-phosphonomethoxypropyl) derivatives of purine and pyrimidine bases. I. The stepwise approach.* Coll Czech Chem Commun 1995, 60: 1196-212.

2. Holy, A., Dvorakova, H., DeClerq, E., Balzarini, J.(Inst. Org. Chem. Biochem. Acad Sci. Czech Republic; Gilead Sci., Inc.). *Antiretroviral enantiomeric nucleoside analogs.* EP 654037, JP 96503927, WO 9403467.

3. Holy, A., Dvorakova, H., Masojidkova, M. *Synthesis of enantiomeric N-(2-phosphonomethoxypropyl) derivatives of purine and pyrimidine bases. II. The synthon approach.* Coll Czech Chem Commun 1995, 60: 1390-409.

4. Arimilli, M.N., Cundy, K.C., Dougherty, J.P., Kim, C.U., Oliyai, R., Stella, V. (Gilead Sci., Inc.). *Nucleotide analogs.* WO 9804569.

5. Schultze, L.M., Chapman, H.H., Dubree, N.J.P. et al. *Practical synthesis of the anti-HIV drug, PMPA.* Tetrahedron Lett 1998, 39:1853-6.

6. Arimilli, M.N., Kim, C.U., Dougherty, J., Mulato, A., Oliyai, R., Shaw, J.P., Cundy, K.C., Bischofberger, N. *Synthesis, in vitro biological evaluation and oral bioavailability of 9-[2-(phosphonomethoxy)propyl]adenine (PMPA) prodrugs.* Antivir Chem Chemother 1997, 8: 557-64.

7. Balzarini, J., Holy, A., Jindrich, J., Naesens, L., Snoeck, R., Schols, D., De Clercq, E. *Differential antiherpesvirus and antiretrovirus effects of the (S) and (R) enantiomers of acyclic nucleoside phosphonates: Potent and selective in vitro and in vivo antiretrovirus activities of (R)-9-(2-phosphonomethoxypropyl)-2,6-diaminopurine.* Antimicrob Agents Chemother 1993, 37: 332-8.

8. Balzarini, J., Aquaro, S., Perno, C.-F., Witvrouw, M., Holy, A., De Clercq, E. *Activity of the (R)-enantiomers of 9-(2-phosphonylmethoxypropyl)-adenine and 9-(2-phosphonylmethoxypropyl)-2,6-diaminopurine against human immunodeficiency virus in different human cell systems.* Biochem Biophys Res Commun 1996, 219: 337-41.

9. Balzarini, J., Vahlenkamp, T., Egberink, H., Hartmann, K., Witvrouw, M., Pannecouque, C., Casara, P., Navé, J.-F., De Clercq, E. *Antiretroviral activities of acyclic nucleoside phosphonates [9-(2-phosphonylmethoxyethyl)adenine, 9-(2-phosphonylmethoxyethyl)guanine, (R)-9-(2-phosphonylmethoxypropyl)adenine, and MDL 74, 968] in cell cultures and murine sarcoma virus-infected newborn NMRI mice.* Antimicrob Agents Chemother 1997, 41: 611-6.

10. Robbins, B.L., Srinivas, R.V., Kim, C., Bischofberger, N., Fridland, A. *Anti-human immunodeficiency virus activity and cellular metabolism of a potential prodrug of the acyclic nucleoside phosphonate 9-R-(2-phosphonomethoxypropyl)adenine (PMPA), bis(isopropyloxymethylcarbonyl)PMPA.* Antimicrob Agents Chemother 1998, 42: 612-7.

11. Srinivas, R.V., Fridland, A. *Antiviral activities of 9-R-2-phosphonomethoxypropyl adenine (PMPA) and bis(isopropyloxymethylcarbonyl)PMPA against various drug-resistant human immunodeficiency virus strains.* Antimicrob Agents Chemother 1998, 42: 1484-7.

12. Xiong, X.F., Chen, M.S. *Inhibition of human cytomegalovirus DNA polymerase by PMEA and PMPA diphosphates.* 36th Intersci Conf Antimicrob Agents Chemother (Sept 15-18, New Orleans) 1996, Abst H29.

13. Zidek, Z., Holy, A., Frankova, D. *Immunomodulatory properties of antiviral acyclic nucleotide analogues: Cytokine stimulatory and nitric oxide costimulatory effects.* Int J Immunopharmacol 1997, 19: 587-97.

14. Zidek, Z., Holy, A., Frankova, D. *Antiretroviral agent (R)-9-(2-phosphonomethoxypropyl)adenine stimulates cytokine and nitric oxide production.* Eur J Pharmacol 1997, 331: 245-52.

15. Suruga, Y., Makino, M., Okada, Y., Tanaka, H., De Clercq, E., Baba, M. *Prevention of murine AIDS development by (R)-9-(2-phosphonylmethoxypropyl)adenine.* J Acquir Immune Defic Syndr Hum Retrovirol 1998, 18: 316-22.

16. Hoover, E.A., Myles, M.H., Ebner, J.P., Black, R.J., Bischofberger, N. *Efficacy of 9-(2-phosphonylmethoxypropyl)-adenine for therapy of acute feline immunodeficiency virus infection.* Antivir Res 1998, 37(3): Abst 60.

17. Tsai, C.-C., Follis, K.E., Beck, T.W., Sabo, A., Bischofberger, N., Dailey, P.J. *Effects of (R)-9-(2-phosphonylmethoxypropyl)-adenine monotherapy on chronic SIV infection in macaques.* AIDS Res Hum Retroviruses 1997, 13: 707-12.

18. Bischofberger, N., Tsai, C.-C., Follis, K.E., Sabo, A., Grant, R.F., Beck, T.W., Dailey, P.J., Black, R. *Antiviral efficacy of PMPA in macaques chronically infected with SIV.* Antivir Res 1996, 30(1): Abst 81.

19. Tsai, C.-C., Follis, K.E., Sabo, A., Beck, T.W., Grant, R.F., Bischofberger, N., Benveniste, R.E., Black, R. *Prevention of SIV infection in macaques by (R)-9-(2-phosphonylmethoxypropyl)-adenine.* Science 1995, 270: 1197-9.

20. Van Rompay, K.K.A., Marthas, M.L., Lifson, J.D., Berardi, C.J., Vasquez, G.M., Agatep, E., Dehqanzada, Z.A., Cundy, K.C., Bischofberger, N., Pedersen, N.C. *Administration of 9-[2-(phosphonomethoxy)propyl]adenine (PMPA) for prevention of perinatal simian immunodeficiency virus infection in rhesus macaques.* AIDS Res Hum Retroviruses 1998, 14: 761-73.

21. Van Rompay, K.K.A., Cherrington, J.M., Marthas, M.L. et al. *9-[2-(Phosphonomethoxy)propyl]adenine therapy of established simian immunodeficiency virus infection in infant rhesus macaques.* Antimicrob Agents Chemother 1996, 40: 2586-91.

22. Naesens, L., Bischofberger, N., Augustijns, P., Annaert, P., Van den Mooter, G., Arimilli, M.N., Kim, C.U., De Clercq, E. *Antiretroviral efficacy and pharmacokinetics of oral bis(isopropyloxycarbonyloxymethyl)-9-(2-(phosphonylmethoxypropyl)adenine in mice.* Antimicrob Agents Chemother 1998, 42: 1568-73.

23. Fridland, A., Robbins, B.L., Srinivas, R.V., Bischofberger, N. *Antiretroviral activity and metabolism of bis(POC)PMPA, an oral bioavailable prodrug of PMPA.* 5th Conf Retroviruses Opportunistic Infect (Feb 1-5, Chicago) 1998, Abst 630.

24. Fridland, A., Robbins, B.L., Srinivas, R.V., Arimilli, M., Kim, C., Bischofberger, N. *Antiretroviral activity and metabolism of bis(POC)PMPA, an oral bioavailable prodrug of PMPA.* Antivir Res 1997, 34(2): Abst 27.

25. Cundy, K.C., Sueoka, C., Lynch, G.R., Griffin, L., Lee, W.A., Shaw, J.-P. *Pharmacokinetics and bioavailability of the anti-human immunodeficiency virus nucleotide analog 9-[(R)-2-(phosphonomethoxy)propyl]adenine (PMPA) in dogs.* Antimicrob Agents Chemother 1998, 42: 687-90.

26. Naesens, L., Bischofberger, N., Arimilli, M., Kim, C., De Clercq, E. *Anti-retrovirus activity and pharmacokinetics in mice of bis(POC)PMPA, the bis(isopropyloxycarbonyloxymethyl) oral prodrug of PMPA.* Antivir Res 1997, 34(2): Abst 28.

27. Shaw, J.-P., Sueoka, C.M., Oliyai, R., Lee, W.A., Arimilli, M.N., Kim, C.U., Cundy, K.C. *Metabolism and pharmacokinetics of novel oral prodrugs of 9-[(R)-2-(phosphonomethoxy)propyl]-adenine (PMPA) in dogs.* Pharm Res 1997, 14: 1824-9.

28. *Gilead announces presentation of phase I/II study results of oral PMPA for HIV at upcoming medical conference. Once-daily oral PMPA reduced HIV viral load by 1.22 log.* Gilead Sciences, Inc. Press Release Jan 26, 1998.

29. *PMPA.* Gilead Sciences, Inc. Product Pipeline Jul 14, 1998.

30. Deeks, S.G., Barditch-Crovo, P., Lietman, P.S., Collier, A., Safrin, S., Coleman, R., Cundy, K.C., Kahn, J.O. *The safety and efficacy of PMPA prodrug monotherapy: Preliminary results of a phase I/II dose-escalation study.* 5th Conf Retroviruses Opportunistic Infect (Feb 1-5, Chicago) 1998, Abst LB8.

31. Gilead Sciences begins phase II human testing of oral PMPA for treatment of HIV: Combinations including hydroxyurea to be evaluated. Gilead Sciences, Inc. Press Release Sept 4, 1998.

Additional References

Clinical trials. Gilead Sciences, Inc. Web Site Jul 14, 1998.

Gilead Sciences, Inc. Product Pipeline Jul 14, 1998.

Cundy, K.C., Safrin, S., Coleman, R., Jaffe, H.S. *Oral PMPA prodrug: Relationship between clinical pharmacokinetics, safety and anti-HIV activity.* 12th World AIDS Conf (Jun 28-Jul 3, Geneva) 1998, Abst 12211.

Bischofberger, N., Naesens, L., De Clercq, E. et al. *Bis(POC)PMPA, an orally bioavailable prodrug of the antiretroviral agent PMPA.* 4th Conf Retroviruses Opportunistic Infect (Jan 22-26, Washington DC) 1997, Abst.

Bischofberger, N. *PMEA and PMPA, two acyclic nucleotide analogues with potent antiretroviral activity.* 213th ACS Natl Meet (Apr 13-17, San Francisco) 1997, Abst BIOT 055.

Gilead commences human clinical testing of systemic PMPA for HIV. Gilead Sciences, Inc. Press Release Oct 17, 1996.

Gilead's PMPA significantly reduces HIV levels in phase I/II study: Oral version expected to enter clinical trials soon. Gilead Sciences, Inc Press Release Apr 11, 1997.

Gilead Sciences begins human testing of oral PMPA for treatment of HIV: Data from phase I/II study of intravenous PMPA showed significant reduction of HIV. Gilead Sciences, Inc. Press Release May 7, 1997.