Commentary

The clinical potential of the acyclic (and cyclic) nucleoside phosphonates. The magic of the phosphonate bond

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ARTICLE INFO	ABSTRACT
	The use of the acyclic nucleoside phosphonates, starting with (S)-HPMPA as the prototype, yielded three clinically approved antiviral drugs, cidofovir for the treatment of CMV retinitis in AIDS patients, adefovir dipivoxil for the treatment of chronic hepatitis B and tenofovir disoproxil fumarate for the treatment of HIV infections (AIDS) and HBV infections. This era has now grown to many more acyclic (and cyclic) nucleoside phosphonates (such as the "open ring" DAPy and Fd4A phosphonates) and alkoxyalkyl and phosphonoamidate prodrugs thereof, as well as new clinical applications, including new drug combination regimens for the treatment of AIDS, the chemoprophylaxis of HIV infections, and the anticancer potential against some malignant disorders. © 2011 Elsevier Inc. All rights reserved.

1. Introduction

In 2011 it will be exactly 25 years ago when we published in Nature [1] on "a novel selective broad-spectrum anti-DNA virus agent". The name of the compound, (S)-9-(3-hydroxy-2-phosphonylmethoxypropyl)adenine [(S)-HPMPA], was not revealed in the title of the Nature paper. Five years ago, we published on "antiviral treatment is more effective than smallpox vaccination upon lethal monkeypox virus infection", again in Nature [2] without the name of the antiviral compound, (S)-1-(3-hydroxy-2-phosphonyl-methoxypropyl)cytosine [(S)-HPMPC], being unveiled in the title of the paper.

(S)-HPMPA and (S)-HPMPC [3] heralded the advent of a totally new and unique class of antiviral drugs, the acyclic nucleoside phosphonates (ANPs) [4], which all originated from Antonin Holý's Laboratory at IOCB (Institute of Organic Chemistry and Biochemistry, Prague, Czech Republic), and with tenofovir have yielded one of the most successful drugs in the treatment of HIV, as well as HBV, infections (AIDS).

What is so special about the ANPs? How were they discovered? What makes them uniquely different from the other antiviral drugs? Could the ANPs, and particularly tenofovir, cure the disease (i.e. AIDS)? Or could it be used prophylactically to prevent HIV infection? Or are any new, nucleoside phosphonates, whether acyclic or cyclic, forthcoming? How could prodrugs be designed for the nucleoside phosphonates to better reach their target site in the organism? Addressing these questions will open new perspectives for the clinical usefulness of these nucleotide analogues, thereby pertaining to the importance (or "magic") of the phosphonate bound.

2. (S)-HPMPA: the prototype of the acyclic nucleoside phosphonates

The first acyclic nucleoside phosphonate that opened the era of nucleotide analogues as potential antiviral drugs was (S)-HPMPA [1]. The compound could be conceived as the hybrid between the broad-spectrum antiviral agent (S)-9-(2,3-dihydroxypropyl)adenine (DHPA) [5] and the specific anti-DNA virus agent phosphonoformic acid (PFA) (Fig. 1). (S)-HPMPA exhibited activity against a variety of DNA viruses, including herpesviruses [herpes simplex virus type 1 (HSV-1) and type 2 (HSV-2), varicella-zoster virus (VZV), cytomegalovirus (CMV)], poxviruses (i.e. vaccinia virus) and adenoviruses [1], and, as shown later [6,7], hepadnaviruses (i.e. hepatitis B virus (HBV)] as well.

The anti-poxvirus activity of (S)-HPMPA could be markedly increased both *in vitro* and *in vivo* when alkoxyalkyl [i.e. hexadecyloxypropyl (HDP) or octadecyloxyethyl (ODE)] esters were attached to (S)-HPMPA [8,9] (Fig. 1). Similarly, the activity of (S)-HPMPA against CMV [8] and adenovirus [10] could be markedly enhanced when converting (S)-HPMPA into its prodrugs with HDP or ODE (for a current state of the art on the antiviral activity of alkoxyalkyl prodrugs of acyclic nucleoside phosphonates, see [11]).

(S)-HPMPA is virtually inactive against HIV [12], and no antiviral activity has ever been noted with (S)-HPMPA or any other acyclic nucleoside phosphonate(s) against RNA viruses. It is surprising, therefore, that the HDP and ODE esters of (S)-HPMPA would be active (in the 0.4–7 nanomolar range) against HIV-1 [13]

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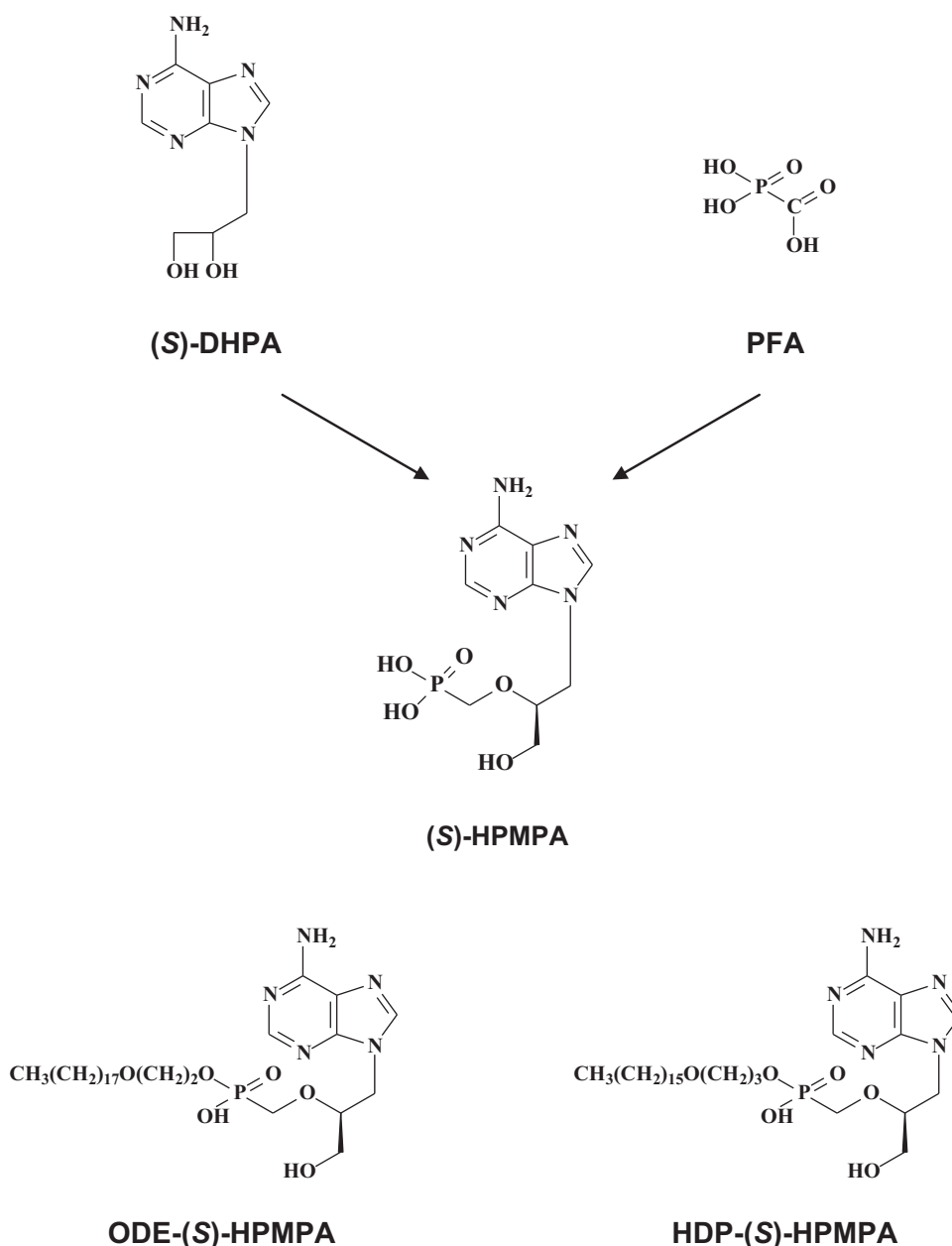
Fig. 1. Structures of (S)-DHPA, PFA, (S)-HPMPA, ODE-(S)-HPMPA and HDP-(S)-HPMPA.

and even more surprising that HDP- and ODE-(S)-HPMPA should be effective against hepatitis C virus (HCV) replication [ODE-(S)-HPMPA at a 50% effective concentration of about 1 μM) [14].

3. (S)-HPMPC: the first acyclic nucleoside phosphonate to be licensed for clinical use

Shortly after (S)-HPMPA, its cytosine counterpart, (S)-HPMPC (cidofovir, Vistide®) (Fig. 2) was reported as a broad-spectrum anti-DNA viral agent, with an activity spectrum similar to that of (S)-HPMPA [3]. The compound was found to be particularly active against CMV [15], and became the first ANP to be approved in 1996 for the treatment of CMV retinitis in AIDS patients. Nowadays, CMV retinitis has virtually disappeared thanks to the successful treatment of AIDS with anti-HIV drugs. This makes that currently cidofovir (Vistide®) is primarily used off-label in the treatment of human papilloma virus (HPV) infections [16], poxvirus infections

(i.e. molluscum contagiosum), adenovirus infections, and polyoma (JC and BK) virus infections [17].

Numerous, albeit anecdotal, case reports have pointed to the efficacy of local intratumoral injection, topical application or even systemic (intravenous) injection of cidofovir in patients with hypopharyngeal papilloma [18], laryngeal papilloma [19], recurrent respiratory papillomatosis [20], plantar warts [21], molluscum contagiosum [22], and orf (ecthyma contagiosum) [23]. As mentioned in the Introduction, cidofovir has also proven more efficacious than smallpox vaccination in the prophylaxis of monkeypox virus infection in monkeys [2].

To increase the oral bioavailability of cidofovir, alkoxyalkyl [i.e. HDP and ODE] esters (Fig. 2) of cidofovir have been developed [11,24]. One of these alkoxyalkyl ester derivatives, i.e. HDP-cidofovir (CMX001) is in clinical development for the therapy and/ or prophylaxis of orthopoxvirus infections [11]. In principle, the alkoxyalkyl esters of cidofovir may be considered for any

(S)-HPMPC

(S)-cHPMPC

ODE-(S)-HPMPC

ODE-(S)-cHPMPC

HDP-(S)-HPMPC
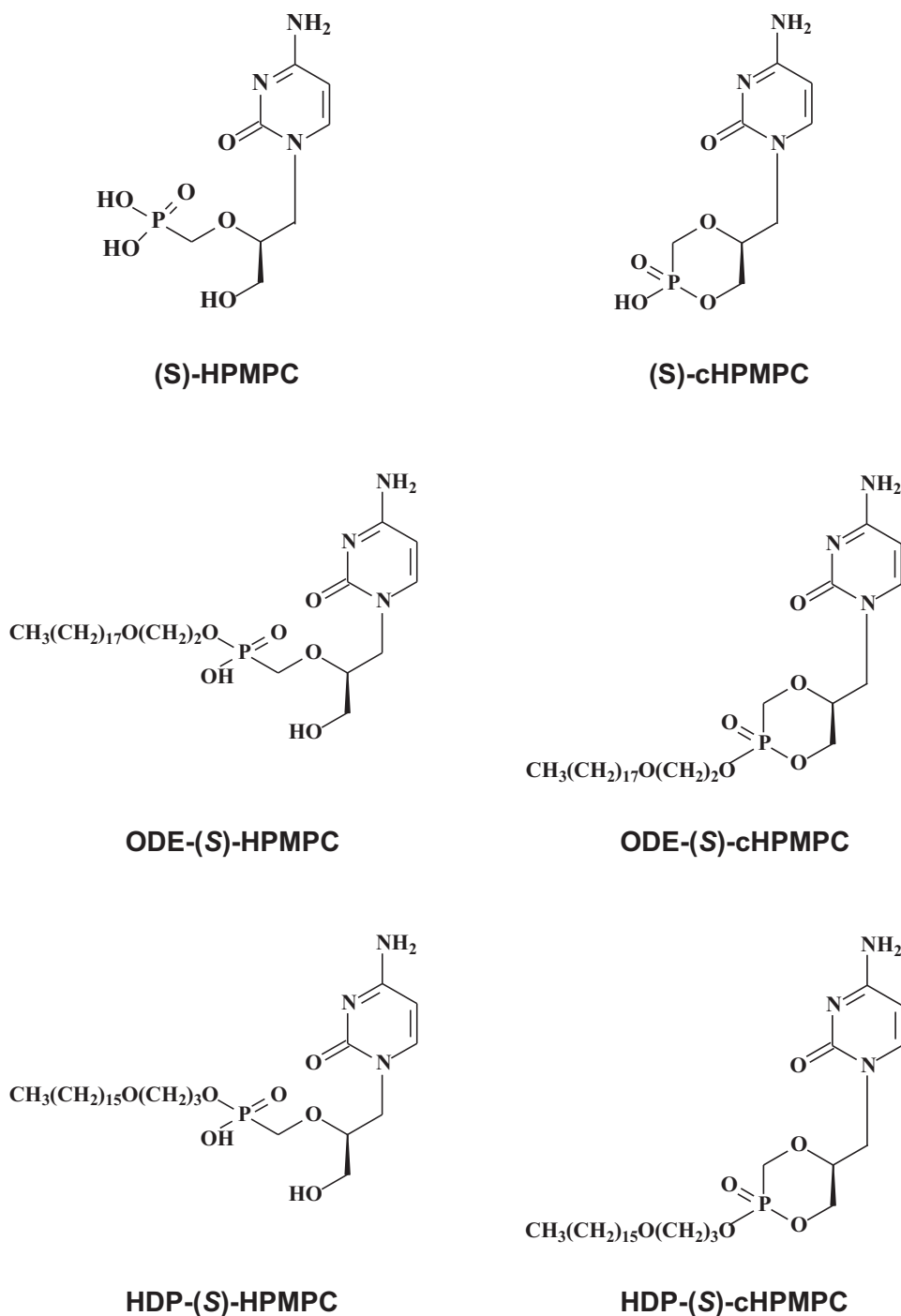
HDP-(S)-cHPMPC

Fig. 2. Structures of (S)-HPMPC, (S)-cHPMPC, ODE-(S)-HPMPC, ODE-(S)-cHPMPC, HDP-(S)-HPMPC and HDP-(S)-cHPMPC.

indication applicable to cidofovir: herpes (HSV, CMV), papilloma-, pox-, adeno- and polyomavirus infections.

4. PMEA: an antiretroviral agent finally licensed for the treatment of hepatitis B

The antiretroviral properties of 9-(2-phosphonylmethoxyethyl)adenine (PMEA) (Fig. 3), which, unlike (S)-HPMPA and (S)-HPMPC, does not contain a chiral carbon center and, therefore, does not exist of (R)- and (S)-enantiomers, were first mentioned by De Clercq et al. [1]. From a mechanistic viewpoint, all acyclic nucleoside phosphonates (ANPs) need two consecutive intracellular phosphorylations, to their diphosphate form, before they can

interact with their target enzyme (DNA polymerase for DNA viruses, reverse transcriptase for retroviruses) as alternate substrates (with respect to dATP, for PMEA diphosphate or (S)-HPMPA diphosphate).

At the DNA polymerase (or reverse transcriptase) level, the ANPs act as obligate chain terminators. For CMV DNA polymerase, two consecutive incorporations of cidofovir [(S)-HPMPC] are required for chain termination to occur [25], while for PMEA, one single molecule incorporated by the viral reverse transcriptase suffices to terminate further chain elongation [26,27].

As for all the ANPs, oral bioavailability of PMEA is limited, which explains why it was converted to its oral prodrug, the bis(pivaloyloxymethyl) ester [28,29] (Fig. 3), which was initially pursued
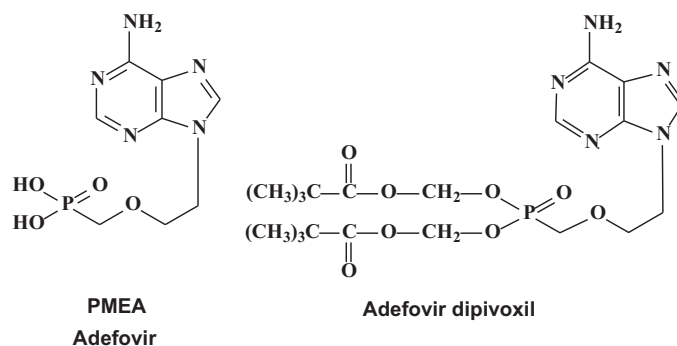
Fig. 3. Structures of PMEA (adefovir) and adefovir dipivoxil.

as an anti-HIV drug. However, at the dosage (62.5 mg or 125 mg per day) required to inhibit HIV replication adefovir dipivoxil [the bis(pivaloyloxymethyl) ester of PMEA] was considered too nephrotoxic to permit long-term (>6 months) use. For the treatment of hepatitis B, however [30,31], the dosage of 10 mg adefovir dipivoxil per day sufficed to obtain a 4 $\log_{10}$, in HBV titer, without any nephrotoxicity, which made the compound, in 2002, licensed for the treatment of hepatitis B.

5. (R)-PMPA and (R)-PMPDAP: potent and selective antiretroviral agents

In 1991 we reported the anti-HIV activity of 9-(2RS)-3-fluoro-2-phosphonylmethoxypropyl derivatives of adenine and 2,6-diaminopurine [32]. Shortly thereafter, in 1993, we showed that this anti-HIV activity resided with the (R)-enantiomers and extended to (R)-PMPA and (R)-PMPDAP (Fig. 4) [12]. In vitro (R)-PMPDAP was about 10-fold more potent than (R)-PMPA against HIV, and, likewise, (R)-PMPDAP was shown to be about 10-fold more potent than (R)-PMPA against HBV [33]. Yet, (R)-PMPA (tenofovir) was developed further as an anti-HIV drug, and later as an anti-HBV drug as well. Its mechanism of action is assumed to be very similar to that of PMEA (adefovir) in that, following intracellular phosphorylation to its diphosphate, tenofovir acts as an (obligate) chain terminator [34] in both the HIV- and HBV-driven reverse transcriptase reaction.

Of crucial importance for the development of (R)-PMPA (tenofovir) were the observations of Tsai et al. [35]: they found that in macaques (R)-PMPA could completely prevent simian immunodeficiency virus (SIV) infection, when administered subcutaneously starting 48 h before, or 4 or 24 h after intravenous SIV inoculation. These startling observation could now, 15 years later, be considered of key value in both the therapeutic and prophylactic usefulness of tenofovir in the control of HIV infections.

6. Tenofovir prodrugs: tenofovir disoproxil fumarate (TDF) and the phosphonoamidate GS-7340

To increase the oral bioavailability of tenofovir, in analogy with the bis(pivaloyloxymethyl) esters of PMEA, the bis(isopropyloxycarbonyloxymethyl) ester of (R)-PMPA was prepared [36,37] and this prodrug (tenofovir disoproxil) was then formulated as the fumarate salt (Fig. 5) to be licensed for clinical use for the treatment of HIV infection (AIDS) in 2001, and for the treatment of HBV infection (chronic hepatitis B) in 2008.

To enhance the delivery of tenofovir into the lymphoid cells (peripheral blood lymphocytes and lymphatic tissues), the alaninyl phenyl ester phosphonoamidate of tenofovir, GS-7340 (Fig. 5) was constructed, and this prodrug of tenofovir showed 1000-fold

enhanced potency against HIV-1 as compared to the parent tenofovir [38] (Fig. 5). It would now seem imperative to determine the clinical value of GS-7340 relative to that of TDF in the therapy and/or prevention of HIV infections.

A third, orally bioavailable, prodrug of tenofovir is represented by CMX-157 (hexadecyloxypropyl-tenofovir) [39], which may have enhanced potency in vitro against NRTI-resistant HIV strains relative to tenofovir [40] and which is in preclinical development for the treatment of HIV infections [11].

7. Cyclic nucleoside phosphonates: PMDTT, PMDTA, Fd4A phosphonate (GS-9148) and its phosphonoamidate prodrug GS-9131

In comparison with the acyclic nucleoside phosphonates, relatively few cyclic nucleoside phosphonates with antiviral potential have been reported, representative examples being the deoxythreosyl phosphonate nucleosides PMDTT and PMDTA [41] (Fig. 6); PMDTT and PMDTA have been reported to be selective anti-HIV agents. However, no follow-up studies on these compounds have so far appeared. Too little is presently known about the activity spectrum (HBV in addition to HIV?), resistance
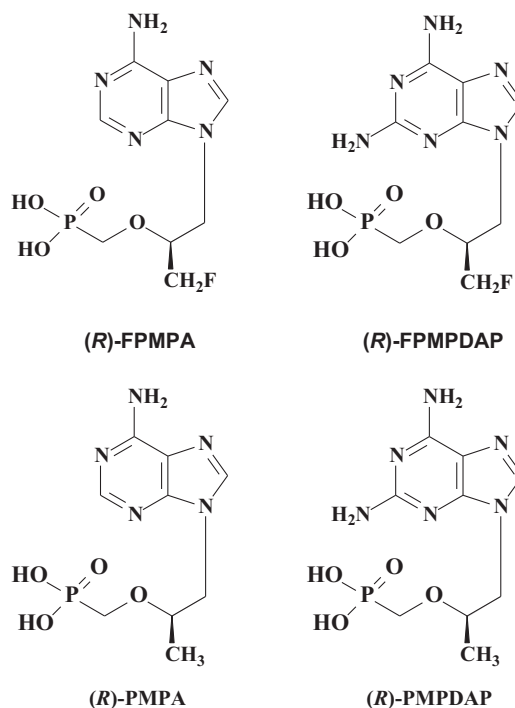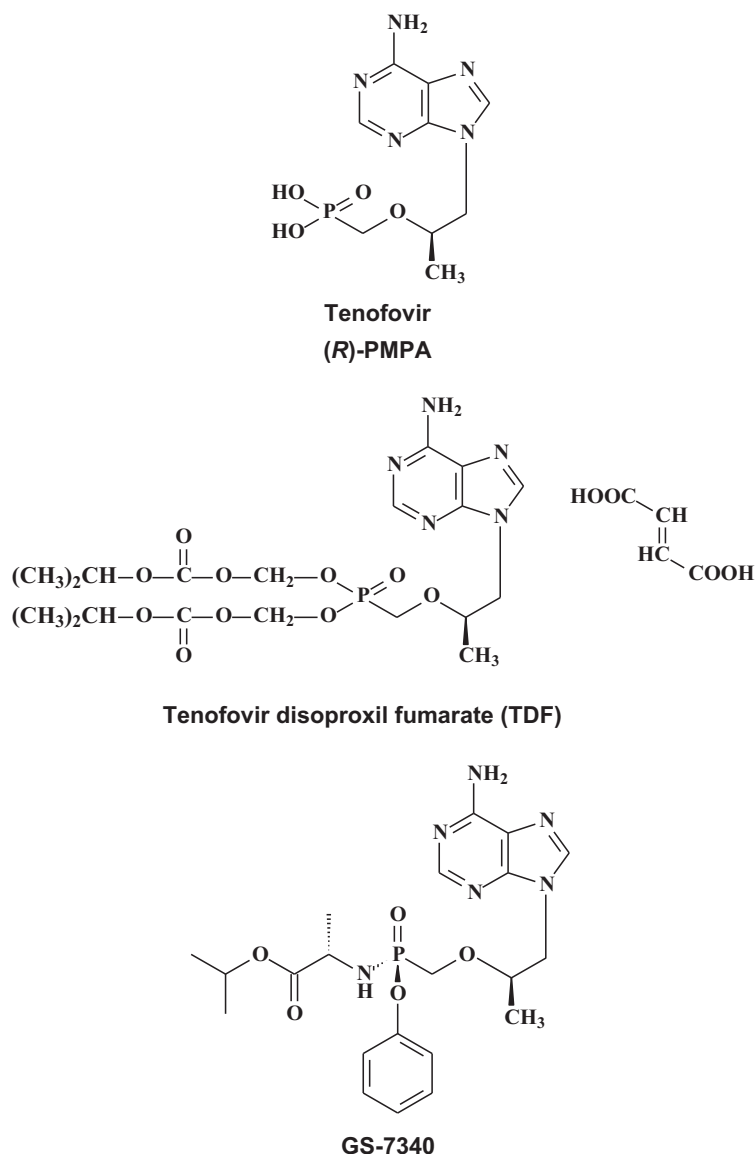

Fig. 4. Structures of (R)-FPMPA, (R)-FPMPDAP, (R)-PMPA and (R)-PMPDAP.

Fig. 5. Structures of (R)-PMPA (tenofovir), tenofovir disoproxil fumarate (TDF) and GS-7340.

profile, and pharmacological, pharmacokinetic and toxicological behavior of this type of compounds to assess their therapeutic potential.

Another cyclic nucleoside phosphonate, the 2′,3′-dideoxy-2′,3′-didehydroadenosine (d4A) phosphonate, was already reported in 1991 [42]. Since this compound is efficiently utilized by mitochondrial DNA polymerase γ, it was suspected to be potentially toxic for the mitochondria [43]. To reduce the risk for mitochondrial toxicity, phosphonomethoxy-2′-fluoro-2′,3′-dideoxy-2′,3′-didehydroadenosine (GS-9148) was synthesized (Fig. 6) [44], and this compound, while designed to reduce mitochondrial toxicity potential, also showed a favorable in vitro resistance profile, retaining activity against a wide range of NRTI resistance mutations including TAMs, K65R, L74V and M184V [44]. GS-9131, the ethylalaninyl phenyl ester phosphonoamidate prodrug of GS-9148, had a 100-fold greater activity in vitro, and allowed for a substantial accumulation and prolonged retention of GS-9148 diphosphate in peripheral lymphocytes in vivo [45]. Lower doses of orally administered GS-9131 are expected to translate into potent clinical antiviral activity, with lower potential for renal toxicity compared to the acyclic nucleoside phosphonates [46]. GS-9131 (Fig. 6), therefore, seems to be an attractive

candidate for clinical development for the therapy and/or prophylaxis of HIV infections.

8. 6-[2-(Phosphonomethoxyalkoxyl]-2,4-diaminopyrimidines (DAPys): pyrimidine derivatives that act as purines

In 2002 we reported on the antiviral activity of a new class of acyclic nucleoside phosphonates, 6-[2-(phosphonomethoxy)alkoxy]pyrimidines [47–49]. This class of compounds (Fig. 7) could actually be divided in two subclasses: first (i), the (R)-HPMPO-DAPy derivatives, which akin to (S)-HPMPC, showed activity against herpes-, adeno-, pox- and papillomaviruses, and, second (ii), the PMEO-DAPy, 5-X-PMEO-DAPy and (R)-PMPO-DAPy derivatives which, akin to PMEA and (R)-PMPA, showed activity against retroviruses (HIV-1, HIV-2) and hepadnaviruses (HBV) [50]. The activity of (R)-HPMPO-DAPy against poxviruses (i.e. camelpox) has in the mean time been confirmed [51], and the 5-methyl-PMEO-DAPy derivative has proven to exhibit pronounced antiretroviral activity in mice infected with Moloney murine sarcoma virus (MSV), but, as to the clinical potential of the DAPys no further information has been forthcoming lately.
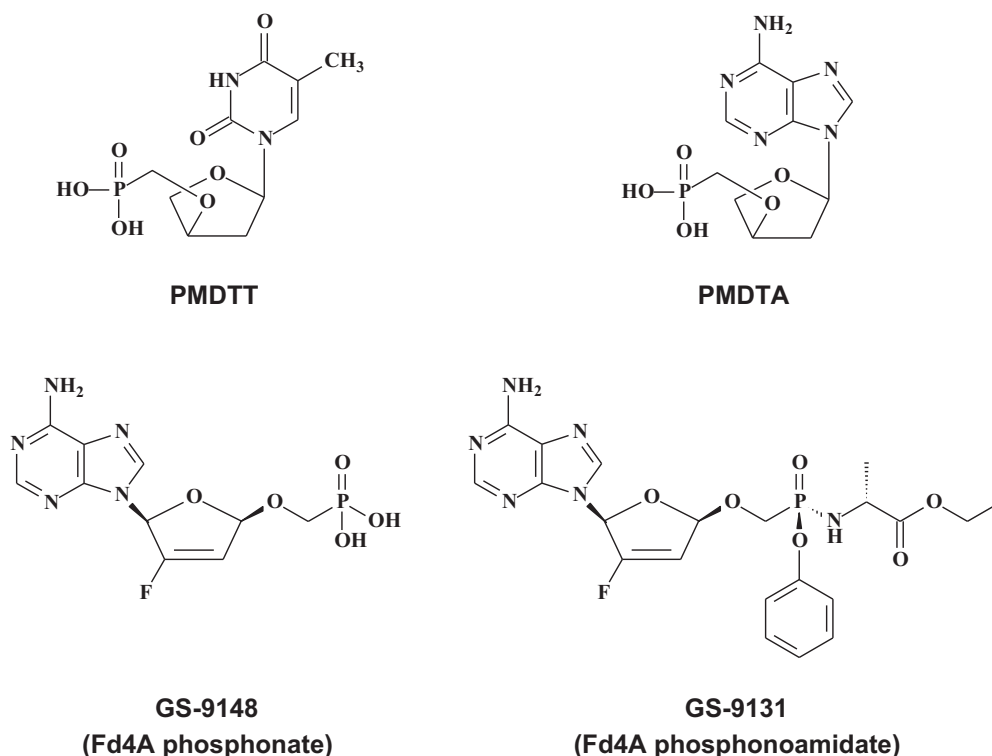
Fig. 6. Structures of PMDTT, PMDTA, GS-9148 and GS-9131.

As could be easily predicted from their chemical structure (Fig. 7), the DAPys behave as purine mimetics [52]; at least they do so in the reverse transcriptase reaction. The "open", incomplete purine ring of the DAPys would allow the canonical Watson–Crick base pairing of PMEO-DAPy with thymine. This canonical Watson–Crick base pairing requires hydrogen bonding between the C_4–NH_2 of DAPy and the C_4–O of thymine, and between the N_3–H of thymine and the N_3 of DAPy, but according to the Watson–Crick base pairing rules, there is a third hydrogen bonding possible, that is between the C_2–O of thymine and the C_2–NH_2 of DAPy (equivalent to the hydrogen bonding between the C_2–O of cytosine and the C_2–NH_2 of guanine in the canonical Watson–Crick base pairing between cytosine and guanine). This may in the case of the DAPys explain why they are incorporated more efficiently than (R)-PMPA and not excised as efficiently as (R)-PMPA. Whether this "third" hydrogen bond translates into a better therapeutic and/or resistance profile of PMEO-DAPy and (R)-PMPA-DAPy, as compared to PMEA and (R)-PMPA, respectively, is an intriguing possibility, worthy of further exploration.

Much about the therapeutic (or prophylactic) usefulness of the DAPy derivatives still has to be revealed. The superiority of (R)-HPMPO-DAPy, like that of (S)-HPMPC, over vaccination in preventing monkeypox in monkeys is but one example [2]. Also, the impact of esterification of the DAPy derivatives [(R)-HPMPO-DAPy, PMEO-DAPy and (R)-PMPA-DAPy] with alkoxyalkyls such as HDP or ODE should be further examined.

9. (S)-HPMPC-5-aza (and its alkoxyalkyl prodrugs), or the effect of replacing a pyrimidine by a triazine moiety

Replacing the pyrimidine ring in (S)-HPMPC by a triazine ring (Fig. 8) resulted in the formation of (S)-HPMPC-5-aza, which exhibited activity against DNA viruses that was comparable to, or even better than, that of the parent compound [53]. This antiviral activity could be further enhanced by introduction of alkoxyalkyl groups, the most active being the hexadecyloxyethyl (HDE) ester (Fig. 8) [54]. HDE-(S)-HPMPC-5-aza should be further explored for its therapeutic potential in the treatment of those virus infections that are sensitive to cidofovir, in particular CMV, HSV, HPV, adeno- and poxvirus infections.

The (S)-HPMPC-5-aza derivatives may be advantageous over (S)-HPMPC and its derivatives, since the intracellular metabolism of (S)-HPMPC-5-aza is different from that of (S)-HPMPC, in that it has a lower sensitivity to catabolic deamination and higher rate of phosphorylation and DNA incorporation [55]. This may possibly be reflected by a better therapeutic potential, in the clinical setting, in the treatment of certain virus infections. In addition, (S)-HPMPC-5-aza can be readily decomposed to the N-formylguanidine and further onto the carbamoylguanidine derivatives which do not exhibit either antiviral activity or cytotoxicity. This rapid decomposition may limit the nephrotoxic potential [56], which could make it advantageous over cidofovir under conditions where nephrotoxicity may be a limiting factor.

10. The anticancer potential of acyclic nucleoside phosphonates, a barely explored therapeutic opportunity

In earlier studies we have pointed to the antitumor activity of both PMEA, its 2,6-diaminopurine analogue (PMEDAP) and the N^6-cyclopropyl derivative thereof (cPrPMEDAP) [57,58], and (S)-HPMPC [59–61]. These antitumor effects were not further explored, however. Instead, two phosphonoamidate prodrugs of cPrPMEDAP, namely GS-9191 and GS-9219, have been further pursued for their anticancer potential: (i) GS-9191 (Fig. 9) (L-phenylalanine,N,N′-[[[2-[2-amino-6-(cyclopropylamino)-9H-purin-9-yl]ethoxy]methyl]-phos-phinylidene]bis-,bis(2-methylpropyl) ester), as a topical prodrug of PMEG [9-(2-phosphonylmethoxyethyl)guanine] for the local treatment of HPV lesions [62] and (ii) GS-9219 (Fig. 9) (diethyl
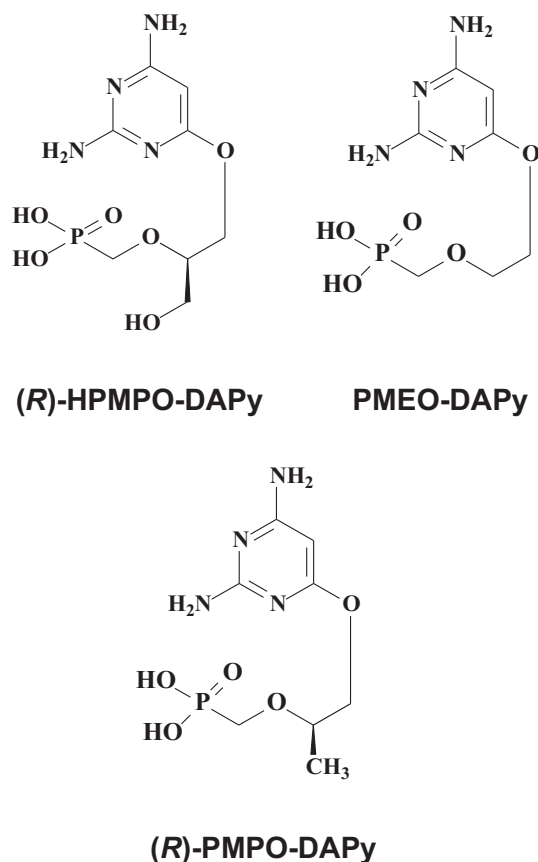
(R)-HPMPO-DAPy   **PMEO-DAPy**

(R)-PMPO-DAPy

Fig. 7. Structures of (R)-HPMPO-DAPy, PMEO-DAPy and (R)-PMPO-DAPy.

N,N′-[({2-[2-amino-6-(cyclopropylamino)-9H-purin-9-yl]ethoxy}-methyl)-phosphonoyl]di-L-alaninate), as an intravenous prodrug for the treatment of non-Hodgkin's lymphoma (NHL) in dogs [63]. In both cases, the active cytotoxic agent is PMEG (first described by De Clercq et al. [3]) which, like all other ANPs, has to be converted to its diphosphate, PMEGpp, before being able to interact, as chain terminator, in the DNA polymerase reaction. Thus, in both cases [(i) and (ii)], GS-9191 and GS-9219 act as pro-prodrugs (or double prodrugs) of PMEG before generating PMEG diphosphate as the ultimate metabolite to unleash the cytotoxic activity. In both cases, the phosphonoamidate prodrugs are intended to direct the PMEG prodrug, cPrPMEDAP to the target tumor cells. The ultimate clinical potential of GS-9191 and 9219, in the (local) treatment of HPV lesions and systemic treatment of NHL, respectively, remains subject of further study.

11. Intracellular conversion of acyclic (and cyclic) nucleoside phosphonates to their diphosphate active metabolite

To exert their antimetabolic (either antiviral or antitumor) activity, all nucleoside phosphonates have to undergo two intracellular phosphorylations, i.e. (S)-HPMPC to (S)-HPMPCpp, PMEA to PMEApp, (R)-PMPA to (R)-PMPApp, and PMEG to PMEGpp (Fig. 10), before the latter can compete with the natural substrate, i.e. dCTP, dATP or dGTP, respectively, at the level of the target enzyme(s), the viral DNA polymerase for (S)-HPMPCpp, the reverse transcriptase for PMEApp and (R)-PMPApp, and cellular DNA polymerases for PMEGpp (Fig. 11). In principle, all the nucleoside phosphonates will act as obligatory chain terminators, although for (S)-HPMPC, two consecutive incorporations may be required before further DNA chain elongation is shut off [25]. Both (S)-HPMPC and (S)-HPMPA can be incorporated into the template strand before strongly inhibiting trans-lesion DNA synthesis (in the case of vaccinia virus DNA polymerase) [64]. Emperically, differential activities have been found for the different acyclic nucleoside phosphonates: (i) for (S)-HPMPC, (S)-HPMPA and the 2,6-diaminopurine analogue thereof [(S)-HPMPDAP] against a broad spectrum of DNA viruses, (ii) for PMEA, (R)-PMPA and their 2,6-diaminopurine analogues PMEDAP and (R)-PMPDAP against HIV and HBV; and (iii) for cPrPMEDAP, via PMEG, against tumor cells. The molecular mechanism underlying this differential action is not known.

Resistance mutations to any of the acyclic nucleoside phosphonates have been shown to develop, albeit only slowly and partially. The best known are K65R (in the HIV reverse transcriptase for HIV resistance against tenofovir), and the N236T (in the HBV DNA polymerase for HBV resistance against adefovir). Where resistance patterns of poxviruses against ANPs were examined, they were not considered of sufficient concern to undermine the use of ANPs in the treatment of poxvirus infections [65]. Resistance development may, therefore, not seem a major concern of ANPs, whatever virus involved, HIV, HBV, or DNA viruses in general.

12. Current therapeutic applications of the acyclic nucleoside phosphonates

The current therapeutic applications of ANPs include (i) for (S)-HPMPC (cidofovir, Vistide®), CMV retinitis in AIDS patients, and off-label, HSV resistant to acyclovir, HPV-associated lesions [16], polyoma (JC and BK) virus infections [17], adeno- and poxvirus infections (i.e. molluscum contagiosum, monkeypox, orf); (ii) for PMEA (adefovir dipivoxil): chronic hepatitis B; and (iii) for (R)-PMPA [tenofovir disoproxil fumarate (TDF, Viread®)], HIV and HBV infections. For the treatment of HIV infections (AIDS), TDF is also available in a fixed-dose drug combination with emtricitabine, as
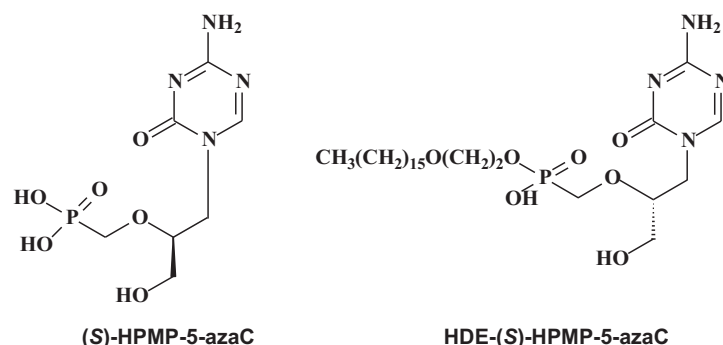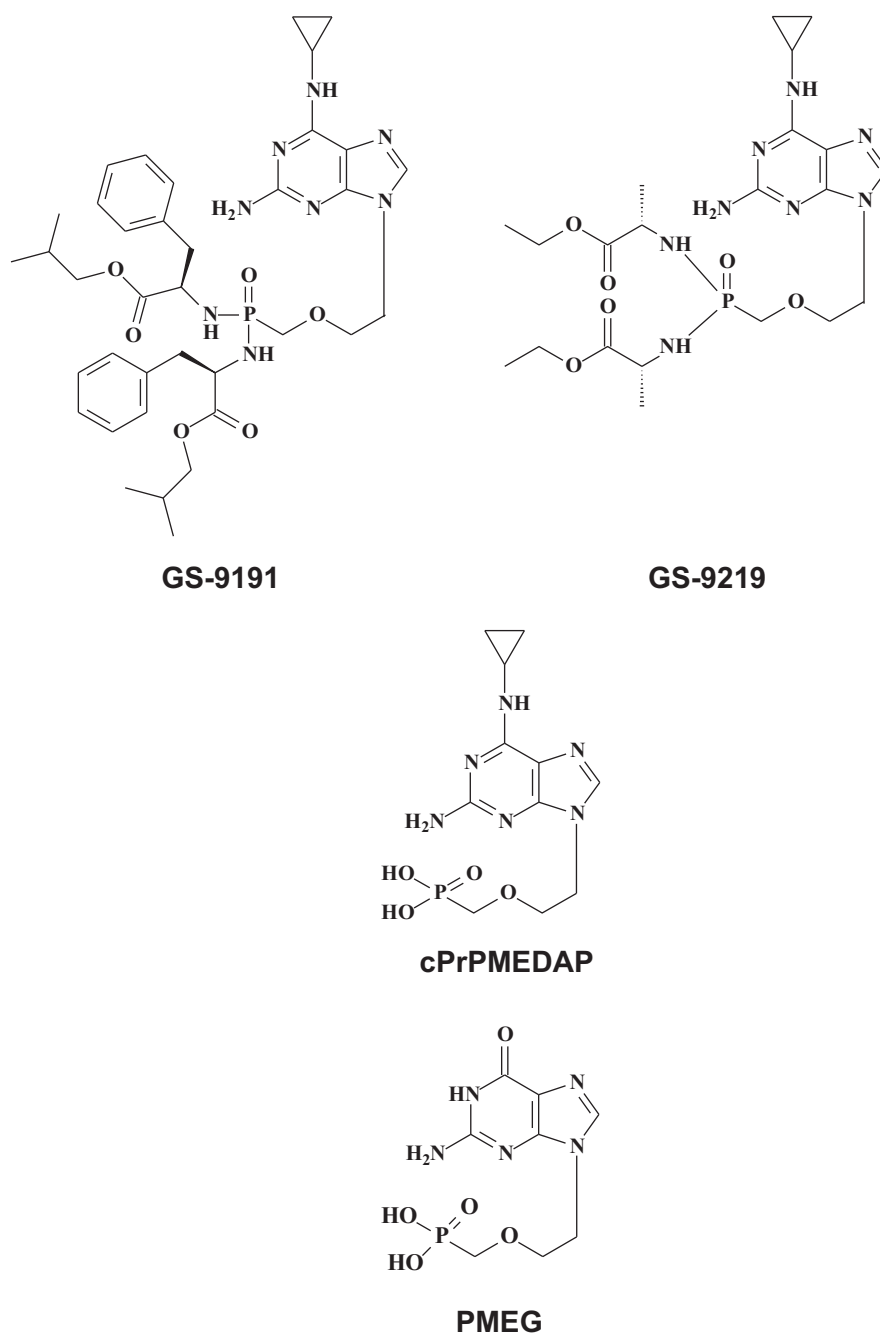

(S)-HPMP-5-azaC   **HDE-(S)-HPMP-5-azaC**

Fig. 8. Structures of (S)-HPMP-5-azaC and HDE-(S)-HPMP-5-azaC.

Fig. 9. Structures of GS-9191, GS-9219, cPrPMEDAP and PMEG.

Truvada®, and a fixed-dose drug combination with both emtri-citabine and efavirenz, as Atripla®.

Forthcoming fixed-dose drug combinations would consist of tenofovir disoproxil fumarate (TDF), emtricitabine and rilpivirine (TMC278), and of tenofovir disoproxil fumarate (TDF), emtricita-bine, Elvitegravir (GS-9137) and cobicistat (GS-9350) (the so-called "Quad" pill).

13. Prophylactic use of tenofovir: prevention of HIV infection

Several experimental observations, (i) prevention of SIV infection in macaques [35], (ii) prophylaxis of intravaginal exposure of pig-tailed macaques to HIV-2 [66], and (iii) protection of infection of newborn macaques against SIV [67] have pointed to the potential value of tenofovir in the prevention of HIV infection, irrespective of the route of transmission (parenteral, vaginal or perinatal). Complete protection from repeated vaginal SHIV (simian–human immunodeficiency virus) exposures was obtained in macaques with a topical gel containing tenofovir alone or with emtricitabine [68], and both *in vitro* and *ex vivo* testing showed tenofovir to be effective as an HIV-1 microbicide [69]. The CAPRISA 004 trial finally showed the effectiveness and safety of vaginal 1% tenofovir gel as an antiretroviral microbicide, for the prevention of HIV infection in women [70].

In 2006 I wrote [71] that based on (i) the original observations of Tsai et al. [35] that SIV infections in macaques could be completely prevented by tenofovir and (ii) the safety efficacy profile established for tenofovir disoproxil fumarate (TDF) in the
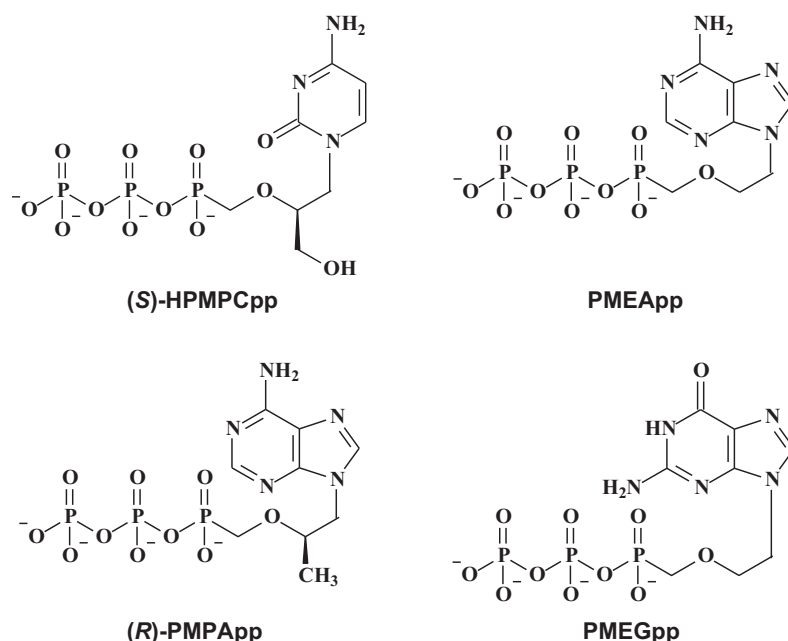
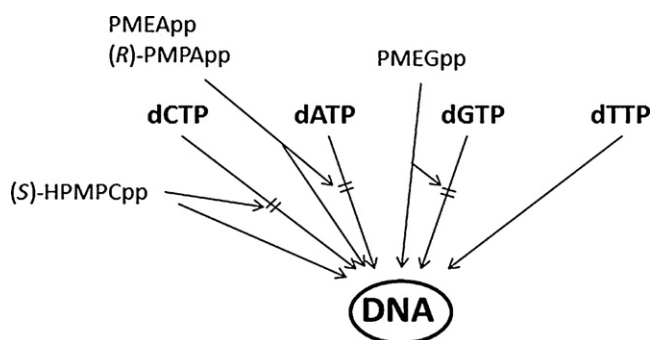**Fig. 10.** Structures of (S)-HPMPCpp, PMEApp, (R)-PMPApp and PMEGpp.

Fig. 11. Modes of action of (S)-HPMPCpp, PMEApp, (R)-PMPApp and PMEGpp.

treatment of AIDS over the past 5-year period (2001–2006) since TDF was approved for clinical use, TDF could be strongly endorsed (as a single daily pill) for the pre- and post-exposure prophylaxis of HIV infections in humans. A number of pre-exposure prophylaxis (PrEP) trials are currently underway with either Viread® or Truvada®, with as target population: intravenous drug users (Thailand), men and women (Botswana), men having sex with men (Peru, Ecuador, USA, South Africa and Brazil) and discordant couples (Kenya and Uganda) [72]. The results of the PrEP with Truvada® for HIV prevention in men who have sex with men have been recently published [73]: these results indicated a 44% reduction in the incidence of HIV, following chemoprophylaxis with Truvada®; this protective effect was additional to the comprehensive package of prevention services offered [73].
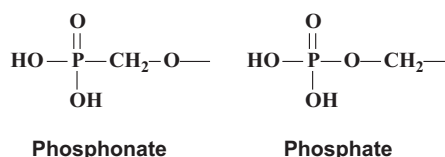

Fig. 12. Phosphonate bond and phosphate bond.

14. Conclusion

Richman et al. recently wrote in Science [74] that the combination therapy for HIV infection represents a triumph for modern medicine, the (only) limitation to the success of this chronic suppressive therapy being the costs involved, the lifelong adherence and the unknown effects of long-term treatment. As an essential component in most of the prevailing combination regimes, tenofovir disoproxil fumarate (TDF) must have contributed to this success.

TDF is but one of the series of either acyclic or cyclic nucleoside phosphonates showing marked antiviral potential. In addition to the "older" acyclic nucleoside phosphonates, (S)-HPMPC, (S)-HPMPA, (S)-HPMPDAP, (R)-PMPA and (R)-PMPDAP, various "newer" acyclic nucleoside phosphonates such as (R)-HPMPO-DAPy, PMEO-DAPy, 5-substituted PMEO-DAPy, (R)-PMEO-DAPy, (S)-HPMPC-5-aza, have been described that yield great potential for the treatment of various DNA virus and retrovirus infections, and so do some cyclic nucleoside phosphonates such as the Fd4A phosphonate GS-9148 and its phosphonoamidate prodrug GS-9131. The PMEG prodrug cPrPMEDAP has potential as an anticancer agent, and for all acyclic nucleoside phosphonates may be made orally bioavailable when esterified by an alkyloxyalkyl.

What all the acyclic and cyclic nucleoside phosphonates have in common is that they contain a phosphonate (PCO) linkage, which unlike the phosphate (POC) linkage (Fig. 12), is not readily hydrolyzed. The phosphonate moiety is isosteric to the phosphate present in the natural nucleotides. The various nucleoside phosphonates, particularly the acyclic nucleoside phosphonates (ANPs), show remarkable differences in their antiviral activity spectrum. Why some ANPs, like (S)-HPMPC, (R)-HPMPO-DAPy and (S)-HPMPC-5-aza are active against a broad range of DNA viruses, and others, like (R)-PMPA, (R)-PMPDAP, (R)-PMPO-DAPy, are specifically active against HIV and HBV, largely remains an enigma linked to the "magic" of the phosphonate bound.

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