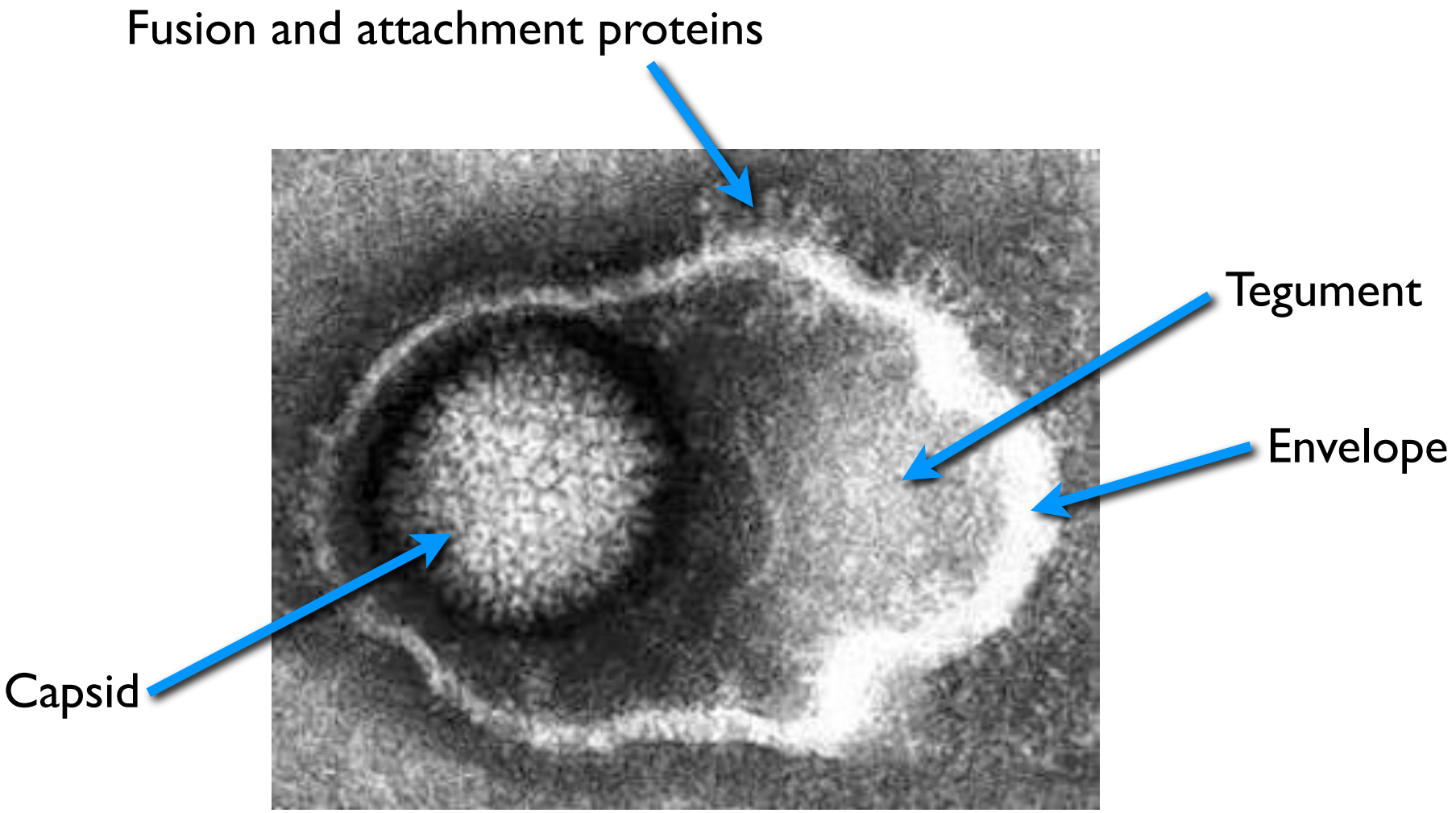
Herpesviruses



Herpesviruses- type and disease

Designation	Common Name	Subfamily	Associated Diseases
HHV-1	HSV-1	Alpha	Oral Herpes (cold sore), Genital Herpes
HHV-2	HSV-2	Alpha	Genital Herpes
HHV-3	VZV	Alpha	Chicken Pox, Shingles
HHV-4	EBV	Gamma	Mononucleosis, Lymphoma, Carcinoma
HHV-5	CMV	Beta	Mononucleosis, Retinitis, Transplant Rejection
HHV-6	HHV-6	Beta	Roseola infantum, Mononucleosis syndrome, Chronic fatigue syndrome, Multiple Sclerosis?
HHV-7	HHV-7	Beta	Roseola infantum, Mononucleosis syndrome?
HHV-8	KSHV	Gamma	Kaposi's Sarcoma

Herpesvirus latency

- After the primary infection, herpesviruses establish latency in the infected host. Once a patient has become infected by herpes virus, the infection remains for life.
- Virus can hide in a quiescent, non-replicating state, often in nerve cells. Controls host's natural cell death mechanisms.
- The latent genome can become activated, in response to various stimulus, to produce infectious virions (e.g. VZV and shingles).
    - Triggers not well-understood:
        - Weakened immune system
        - Other infection
        - Stress
        - Trauma
- Antivirals are not effective against latent "virus"

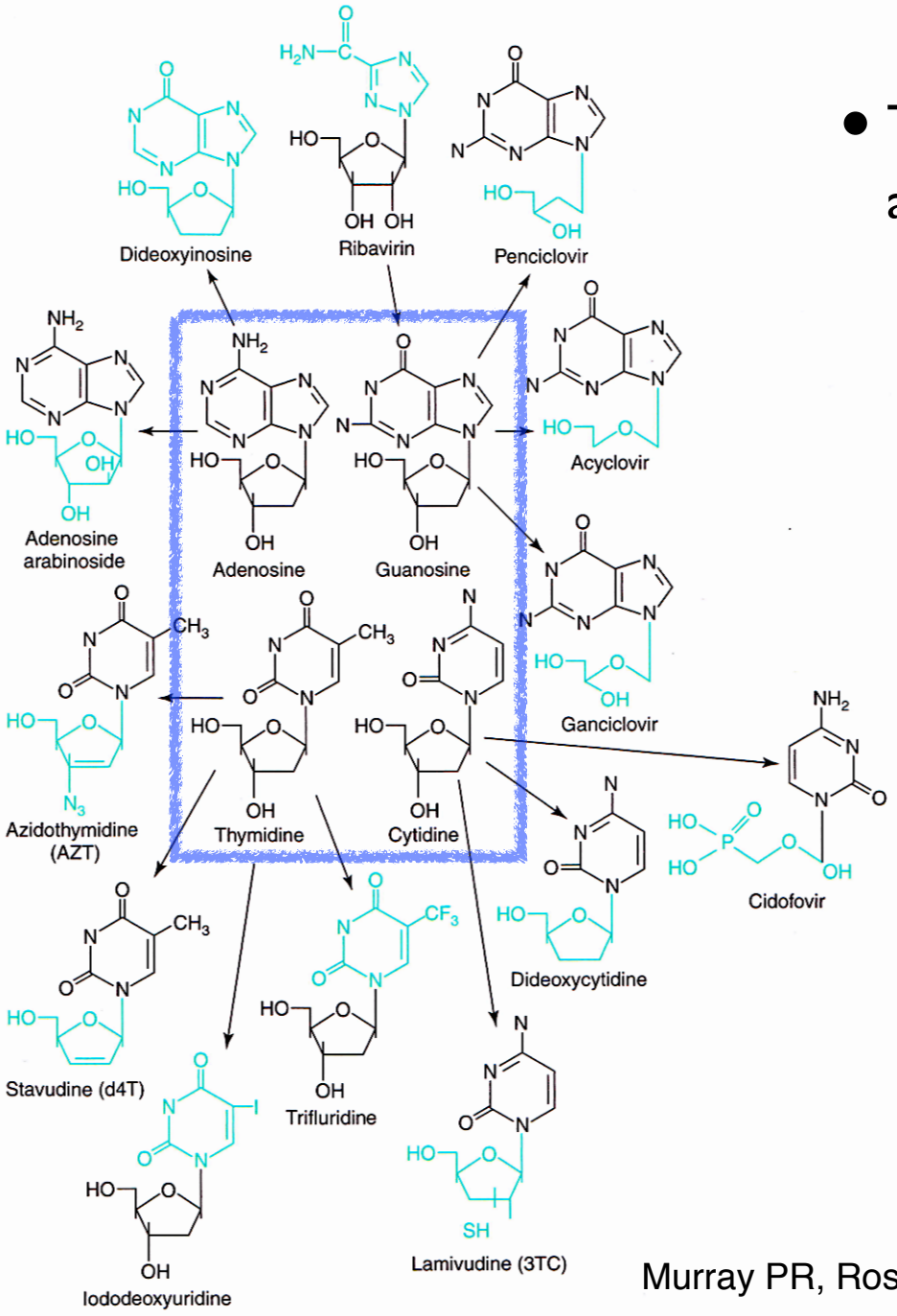
Varicella-Zoster Virus (VZV)

- Varicella virus (a.k.a. Human Herpes Virus-3) causes chicken pox in children
- Latent state, infection not cleared. May reemerge in adults as shingles.
    - Shingles rash can transmit VZV, less contagious than chicken pox rash.
- Approved antivirals for the treatments of VZV:
    - Acyclovir
    - Valacyclovir
    - Famciclovir
- Treatment within 48-72h of shingles eruption for reduction of blisters/pain
    - Reduces later complications such as postherpetic neuralgia (persistent pain for months-years, likely due to nerve damage from VZV)
- A vaccine is available: Zostavax®

Herpesvirus encephalitis

- Caused by HSV-1 (90%) and HSV-2 (10%)
- Retrograde transmission from facial nerves to brain
- 1/500,000 individuals
- Confusion, seizures, fever, elevated WBC in CSF without bacteria or fungi
- Without treatment, fatal in 70% of cases
    - Acyclovir IV treatment

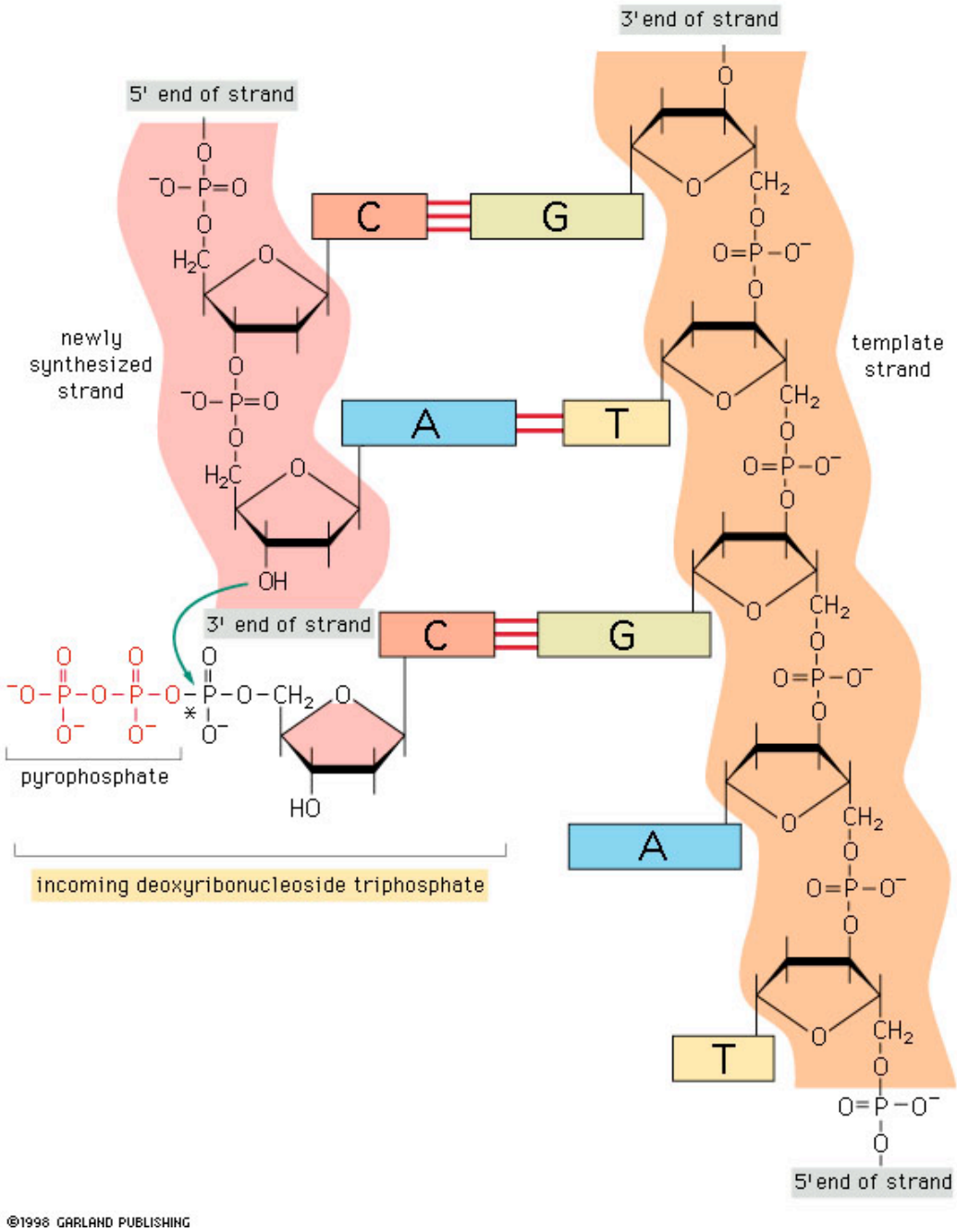
Mechanisms of action: Deoxynucleoside analogs



- Target viral DNA polymerases (herpesvirus) and reverse transcriptase (HIV, hepatitis B)
  - The drugs can selectively inhibit viral genome replication because the viral polymerases are more tolerant to binding incorrect substrate (the drug).
  - The viral polymerases and reverse transcriptases are essential for virus replication and distinct from host enzymes.
  - Activated in cells by viral kinase then by host kinases.

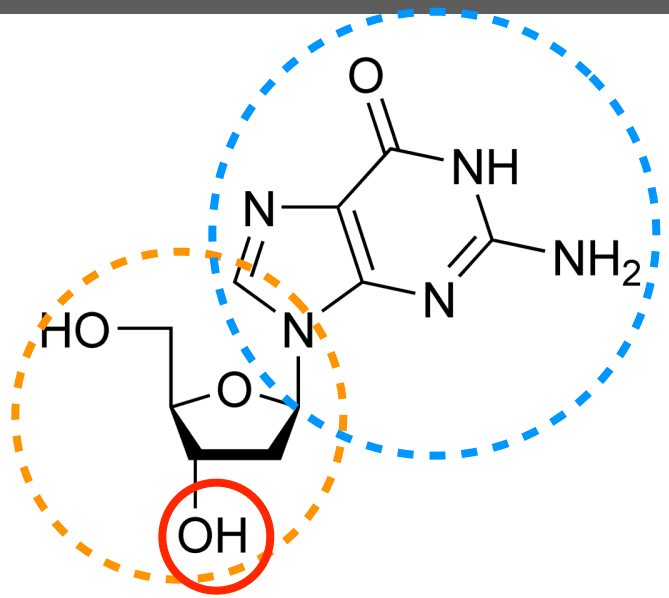
Murray PR, Rosenthal KS, Pfaller MA, (2009) Medical Microbiology 6th Edition

Mechanisms of action: (Deoxy)nucleoside analogs



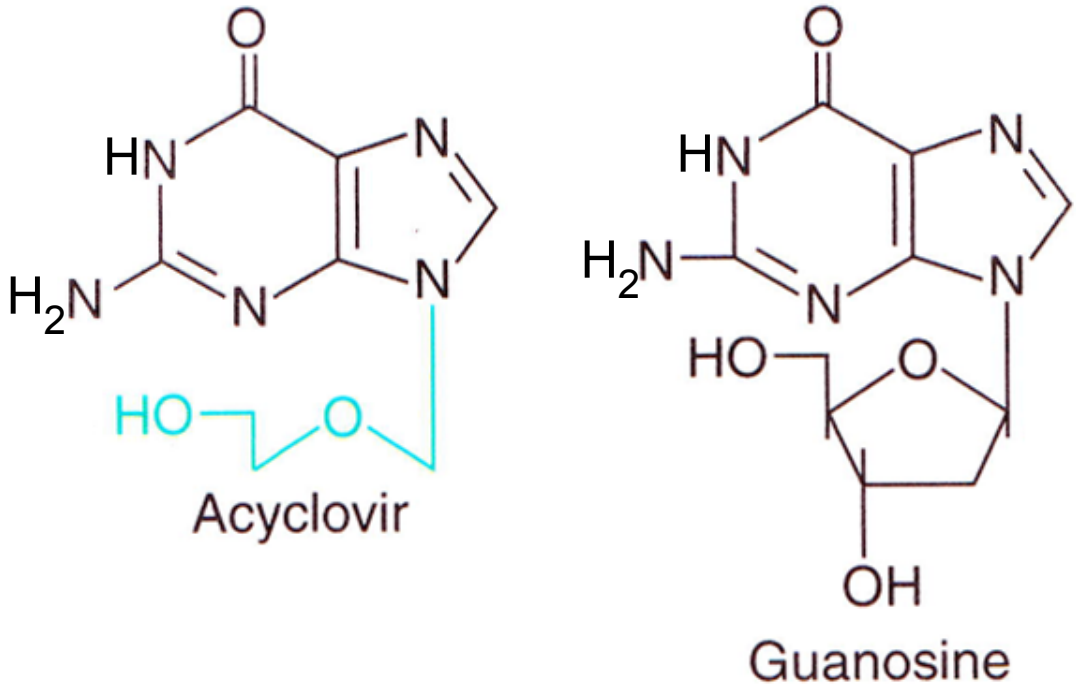
Alberts B et al, *Molecular Biology of the Cell,* 4th Edition

Mechanisms of action: Nucleoside analogs



- Sugar modifications
  - E.g. acyclovir, ganciclovir, valacyclovir, penciclovir, cidofovir, lamivudine (3TC), etc. In some cases, 3'-hydroxyl on "sugar" is missing, and chain termination results
- Base modifications
  - Inactivating mutations due to incorporation into nucleic acid chain (e.g. ribavirin, idoxuridine, trifluridine)
- Viral versus host DNA polymerases: Selectivity of drug for viral polymerase desired (100-1000x greater affinity). Collateral damage if host polymerases affected, toxicity issues (e.g. teratogenic)

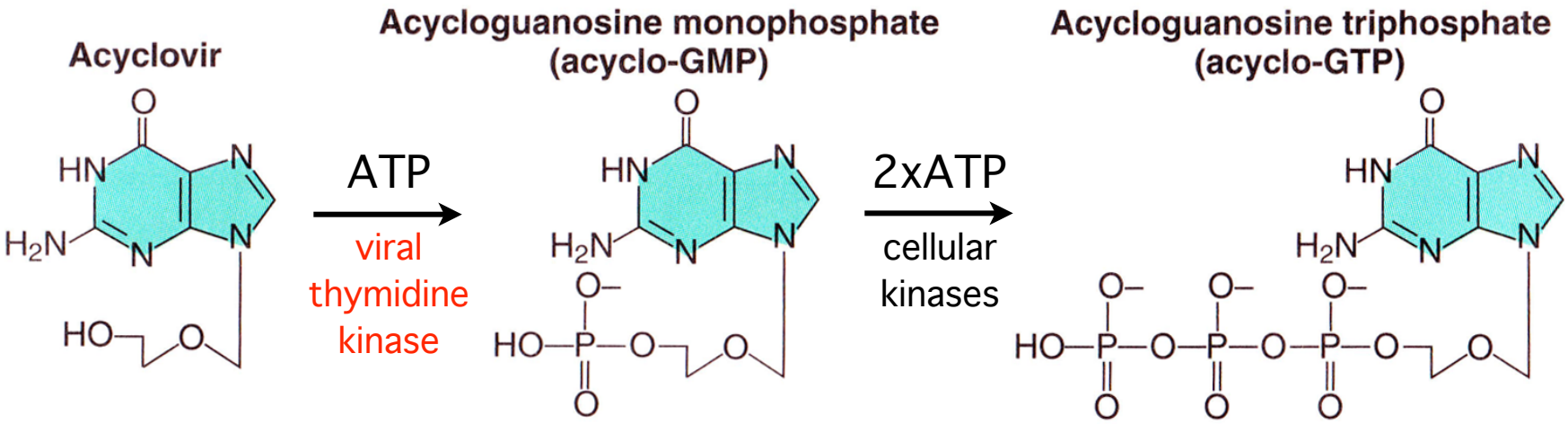
Nucleoside analog example: Acyclovir, a deoxyguanosine analog



- Lack of 3'-hydroxyl on "sugar" makes nucleic acid chain elongation impossible. Chain terminating drug.

Murray PR, Rosenthal KS, Pfaller MA, (2009) Medical Microbiology 6th Edition

Herpesvirus thymidine kinase (TK)

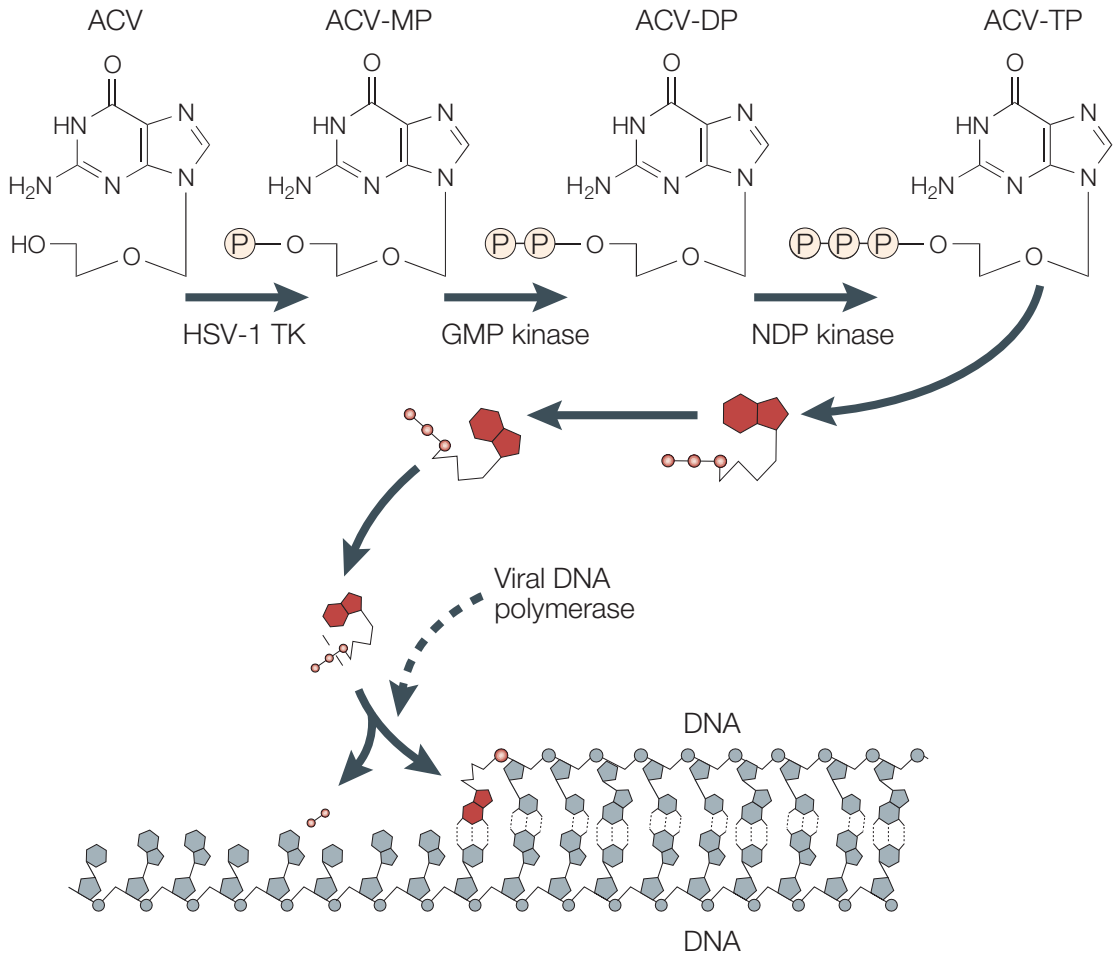


- Herpesviruses encode a kinase ("viral thymidine kinase", TK) that performs the first step in phosphorylation of nucleosides in virally infected cells. Many of the antivirals have a high affinity for TK over host thymidine kinases.

- Selectivity: Because the viral thymidine kinase is absent from uninfected cells nucleoside analog (e.g. acyclovir) is not converted to active form; no initial phosphorylation. Low impact on host DNA synthesis in uninfected cells.

Murray PR, Rosenthal KS, Pfaller MA, (2009) Medical Microbiology 6th Edition

Acyclovir (and other nucleoside analogs) M.o.A.



- Acyclovir is phosphorylated by viral thymidine kinase to acyclovir monophosphate, then by cellular kinases to ACV-triphosphate.
- Acyclovir triphosphate substitutes for guanosine in the growing DNA chain by competing for binding to the viral DNA-polymerase, thus terminating DNA chain proliferation.

De Clercq E, *Nat Rev Drug Discov* **1**:13 (2002)

Acyclovir (and other nucleoside analogs) M.o.A.

- Because the viral thymidine kinase is absent from uninfected cells the acyclovir is not converted to active form; no initial phosphorylation.
- Viral DNA polymerase binds ACV-triphosphate more tightly than cellular DNA polymerase, and hence is selectively inhibited
- Apparently greater uptake of ACV into herpesvirus-infected cells
- Resistance by reduced thymidine kinase activity (ACV cannot get activated; but mutated virus are usually less fit) or DNA polymerase (prevents ACV binding)

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Targeting alpha-herpesviruses: HSV-1, HSV-2, VZV

- Acyclovir
- Valaciclovir
- Penciclovir
- Famciclovir
- Idoxuridine: *HSV-1 only; keratitis; topical opthalmic treatment of cornea*
- Trifluridine: *HSV-1 &2, vaccinia, adenovirus; conjunctivitis, keratitis; topical opthalmic solution*

Acyclovir (Zovirax®, Glaxo-SmithKline)

Spectrum:

- HSV-1: cold sores, keratitis (eye), encephalitis (CNS)
- HSV-2: genital herpes
- Varicella zoster virus (VZV): shingles/chicken pox

Virus	EC50
HSV-1	0.1 μM
HSV-2	0.5 μM
VZV	4.9 μM
CMV	71 μM
EBV	120 μM

Indicated uses:

- Mucosal and cutaneous herpes simplex in immunocompromised patients
- Severe episodes of herpes genitalis
- Herpes simplex encephalitis
- Neonatal herpes virus infections
- VZV (shingles) in immunocompromised patients

Acyclovir

Acyclovir properties:

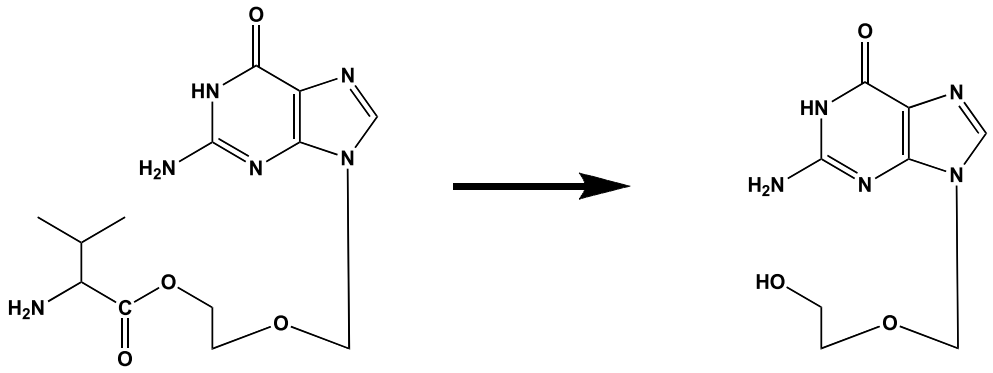
- Poor oral bioavailability (15-20%)
- Topical ointments, creams, PO, and IV
- IV delivery if high concentrations needed (e.g. for encephalitis)
- Up to 91% renal clearance of unmodified drug; adjust dose for renal dysfunction

Adverse effects

- Renal function impairment
- Precipitation of acyclovir in renal tubules (crystalluria); ensure good hydration
- Thrombocytopenic purpura/hemolytic uremia syndrome (rare; immunocompromised)
- Photosensitivity (rash)
- Stinging (30%) with ointment

Nucleoside analog prodrugs

- The active nucleoside analog drugs have very poor oral bioavailability.
- Prodrugs improve bioavailability by increasing transport/uptake and solubility.

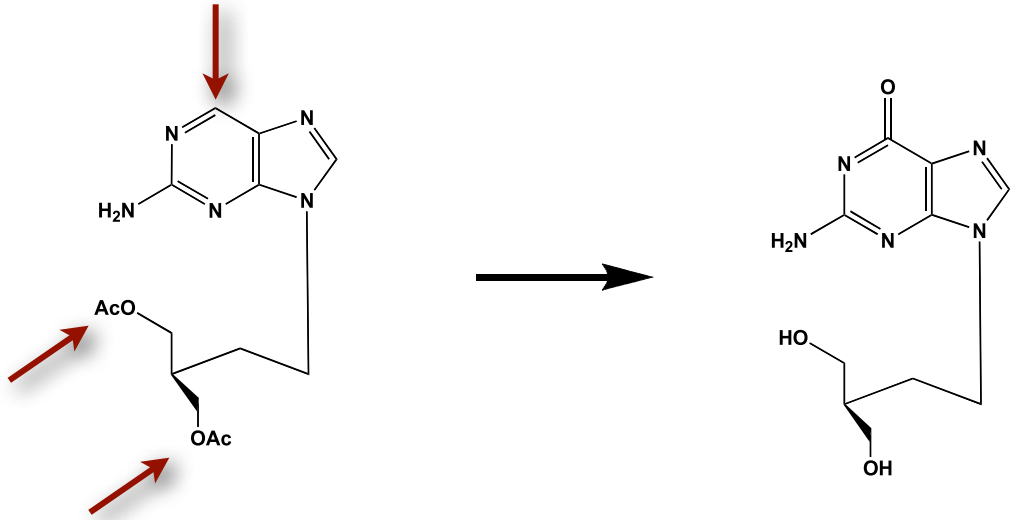


Valacyclovir **Acyclovir**

Valacyclovir (Valtrex®, Glaxo-SmithKline)

- Valine ester prodrug of acyclovir.
- PO; 54% oral bioavailability (vs 10-20% for acyclovir); GI absorption (PEPT1 transporter)
- Metabolized to active acyclovir by esterases in liver/intestines
- Due to prolonged release (activation) of acyclovir, can give fewer doses per day
- Similar indications to acyclovir (ACV); plus chickenpox in immunocompetent children
- Similar adverse reactions to ACV

Famciclovir and Penciclovir

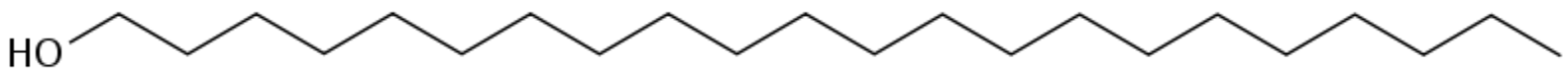


Famciclovir Penciclovir

Famciclovir (Famvir®, Novartis)

- Diacetate ester prodrug of penciclovir
- PO; 77% oral bioavailability (vs 2-5% for penciclovir); absorbed through gut mucosa
  - Penciclovir used only as a topical treatment (1% cream Denavir®)
- Double activation required by carboxylesterases and xanthine oxidase
- Not chain-terminating, ~100x less active than ACV, but intracellular concentrations very high, so still effective
- Similar indications to acyclovir (ACV); often used for VZV/shingles
- Similar adverse reactions to ACV

Docosanol



- Abreva®
- OTC, topical 10% cream indicated for treatment of HSV-1 *labialis* (cold sores)
- Saturated fatty alcohol
- Believed to inhibit membrane fusion between viral envelope and host cell
  - *In vitro* activity against HSV-1&2, RSV, influenza, HIV, other enveloped viruses
- Shortens duration of infection by ~1 day (out of 8-14 days)

Targeting beta-herpesviruses

- Cytomegalovirus
- Drugs with indicated usage for treatment of beta-herpesviruses:
  - Ganciclovir
  - Valganciclovir
  - Cidofovir
  - Foscarnet
  - Fomivirsen

Beta-herpesvirus: Cytomegalovirus (CMV)

- CMV causes few symptoms in children and mild disease in adults. Can be a risk for developing fetus during pregnancy, however (1 in 750 infants are born with or develop some form of disability due to *in utero* CMV exposure-CDC).

- The virus is spread via secretions: saliva, urine, vaginal secretions, semen

- CMV may also be spread by blood transfusion and organ transplant

- Virus elicits humoral and cell-mediated immunity but the infection is not cleared due to virus existing in a latent state or low-level chronic infection

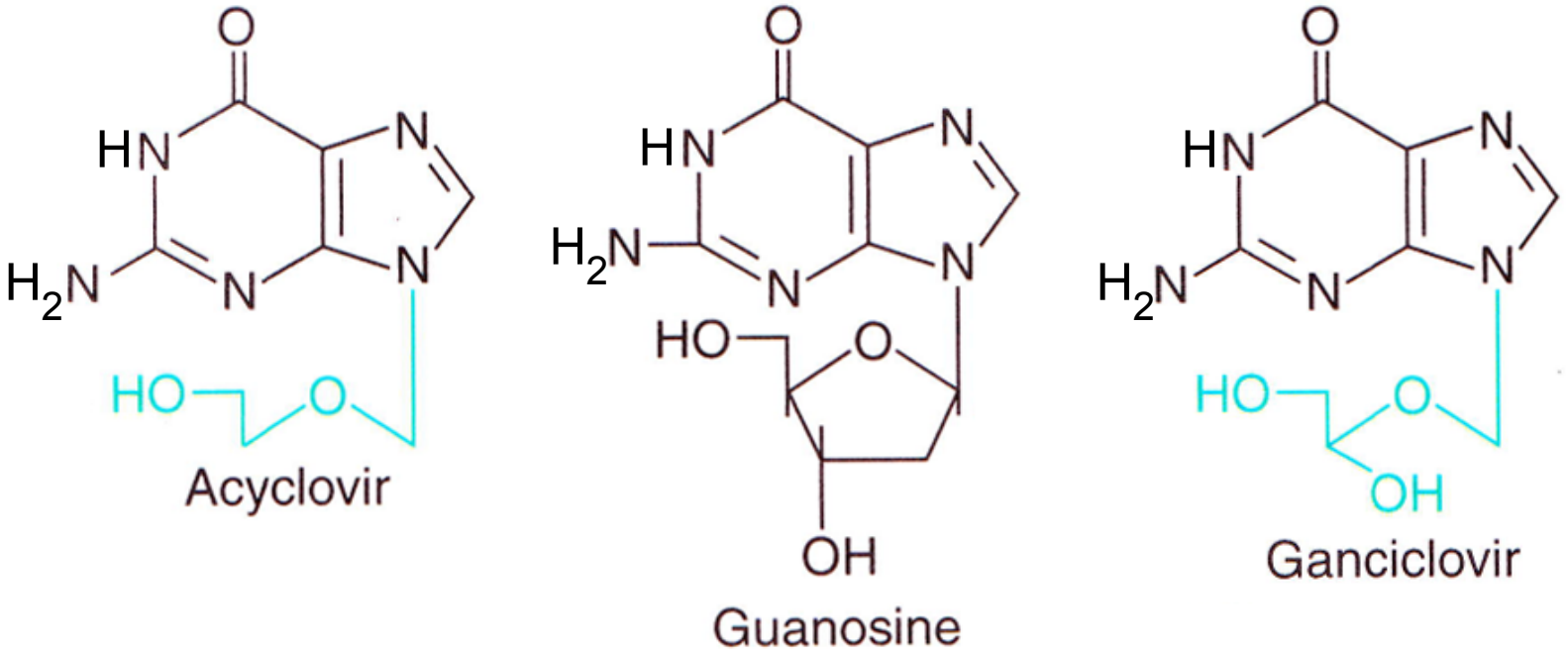
- The virus may reactivate, particularly in cases of:
  - Organ transplant patients
  - Immunosuppressive disease

- CMV-retinitis occurs in up to 15% of all AIDS patients; can lead to blindness

- Also may cause pneumonia, colitis, esophagitis and encephalitis

Ganciclovir

Ganciclovir (Cytovene®, Roche)



- A guanosine analog, similar to acyclovir, but better activity against CMV.
- Ganciclovir is phosphorylated by TK for HSV-1, HSV-2, and VZV, but by a virally-encoded protein kinase (UL97 gene product) for CMV
- Ganciclovir-triphosphate blocks the uptake of dGTP into the growing viral DNA by competing for binding sites. Gets incorporated into viral nucleic acid, but not a chain terminator like acyclovir.

Ganciclovir

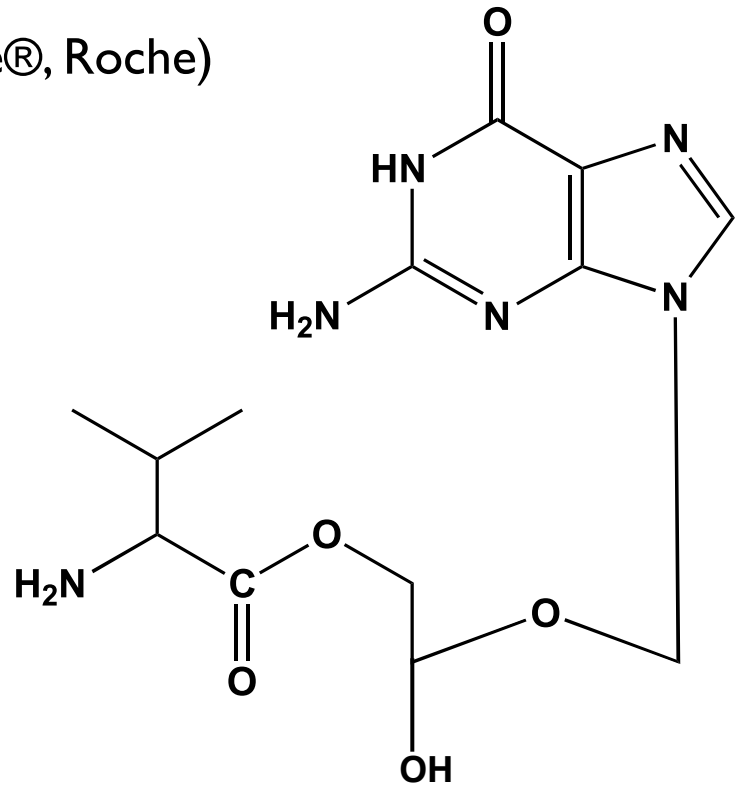
- Broad spectrum anti-herpesvirus activity, but only indicated use is for CMV in immunocompromised patients (retinitis) and as a prophylaxis against CMV infection in organ transplants.

Virus	EC50
HSV-1	0.1 μM
HSV-2	0.1 μM
VZV	2.8 μM
CMV	3.4 μM
EBV	0.1 μM

- IV or intravitreal. Very poor oral bioavailability, 2-7%. Valganciclovir prodrug has ~60%.
- 90-100% renally cleared unmodified.
    - Should be given by slow IV infusion to avoid reaching toxic blood levels of this drug, and dose needs to be adjusted in renal disfunction.
- Granulocytopenia (granulocytes↓), anemia (RBC↓), thrombocytopenia (platelets↓).
- Teratogenic, mutagenic, and carcinogenic in animals. Hypospermia. Pregnancy cat. C.

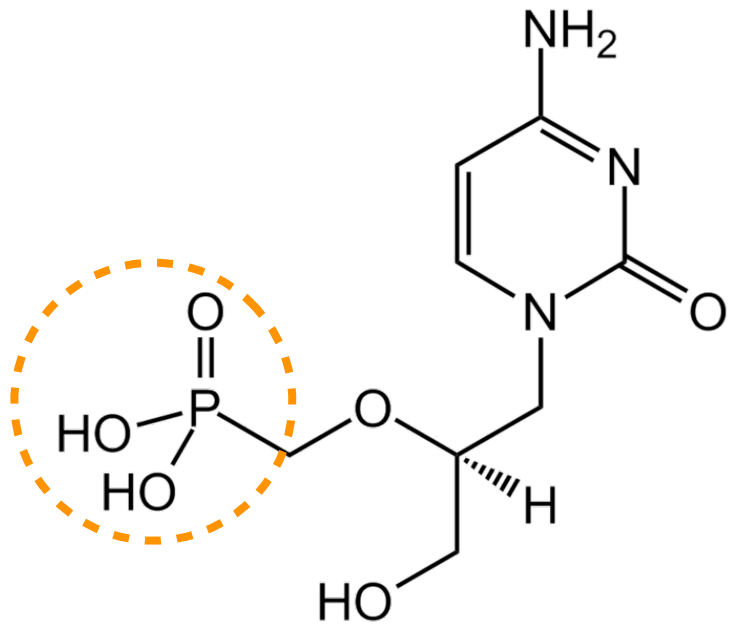
Valganciclovir (Ganciclovir prodrug)

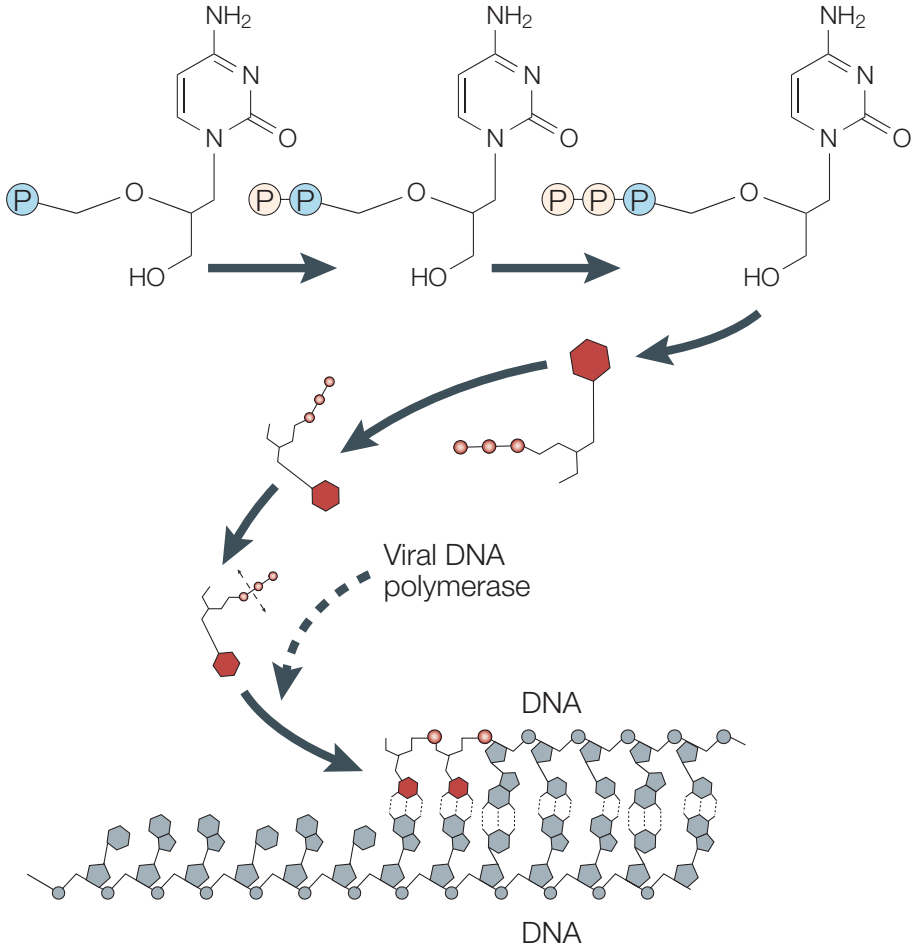
Valganciclovir (Valcyte®, Roche)



- Oral bioavailability ~60% (vs 2-7% for ganciclovir)
- Valyl moiety dramatically increases uptake due to recognition by peptide transporters
- Cellular esterases cleave off the valyl moiety
- Same toxicities as ganciclovir.

Cidofovir: a phosphonated, broad-spectrum nucleotide analog



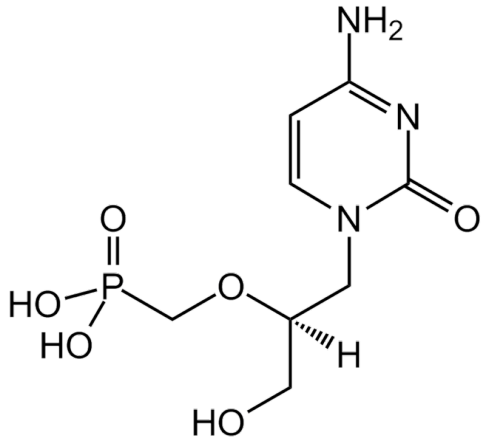


Cidofovir (Vistide® Gilead)

- IV
- Purine nucleoside phosphonate that mimics deoxycytidine monophosphate (dCMP).
- Drug not reliant upon viral TK (only produced by some viruses), spectrum is broader: papillomavirus, herpesvirus, adenovirus, pox virus.
  - Selectivity no longer due to expression of viral TK, only due to preferential binding to viral polymerase over host polymerase (~1000x)

Cidofovir: a broad-spectrum phosphonated-nucleoside analog

- Indicated only for treatment of CMV retinitis in patients with AIDS.

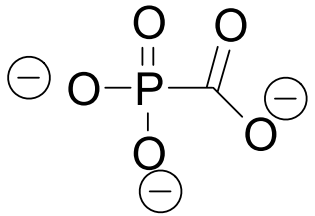


Nephrotoxicity: (taken up by renal proximal tubular cells, induces apoptosis)
- Some cases observed after one or two doses.
- Avoid use with other nephrotoxic drugs.
- Prehydrate the patient well.
- Administer with probenecid to decrease nephrotoxicity.
  - Probenecid competes with cidofovir for OAT-1 transporter in proximal tubular cells, which apoptose due to cidofovir uptake.

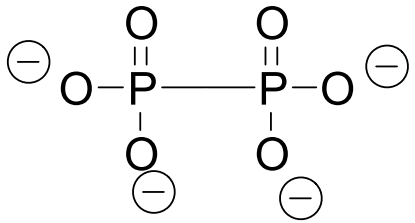
Neutropenia (neutrophils ↓)

Carcinogenic, teratogenic, hypospermia.

Target Herpesvirus DNA polymerase: Foscarnet



Foscarnet



Pyrophosphate

Foscarnet (Foscavir® Astra)

- IV
- An organic analog of inorganic pyrophosphate, which is a product of DNA/RNA replication (PPi comes off the nucleoside triphosphate during polymerization). Foscarnet occupies the PPi site on viral DNA polymerase.
- At working concentrations, low binding to eukaryotic DNA polymerases.
- FDA approved for CMV retinitis and for acyclovir-resistant HSV infections. Usually used after other anti-herpes drugs have failed in immunocompromised patients.

Target alpha-Herpesvirus DNA polymerase: Foscarnet

Adverse effects:

- Nephrotoxic (1/3 of patients): monitor serum creatinine and dose adjust
- Seizures; neurotoxic due to changes in plasma mineral and electrolyte content
    - Forms a chelate with calcium.
    - 10% in AIDS patients.

Herpesvirus drugs summary

- Most are nucleoside analogs that get incorporated into replicating DNA
- Target replicating virus, not active against latent virus
    - Treat and control outbreaks, but does not cure one of the infection

- For alpha-herpes viruses: simplex 1 and simplex 2, VZV:
    - Acyclovir and Valacyclovir (prodrug)
    - Penciclovir and Famciclovir (prodrug)
    - Foscarnet
- For beta-herpes viruses: CMV
    - Ganciclovir and Valganciclovir (prodrug)
    - Cidofovir
    - Foscarnet