Nonclinical Evaluation of Oral Antiviral Agents

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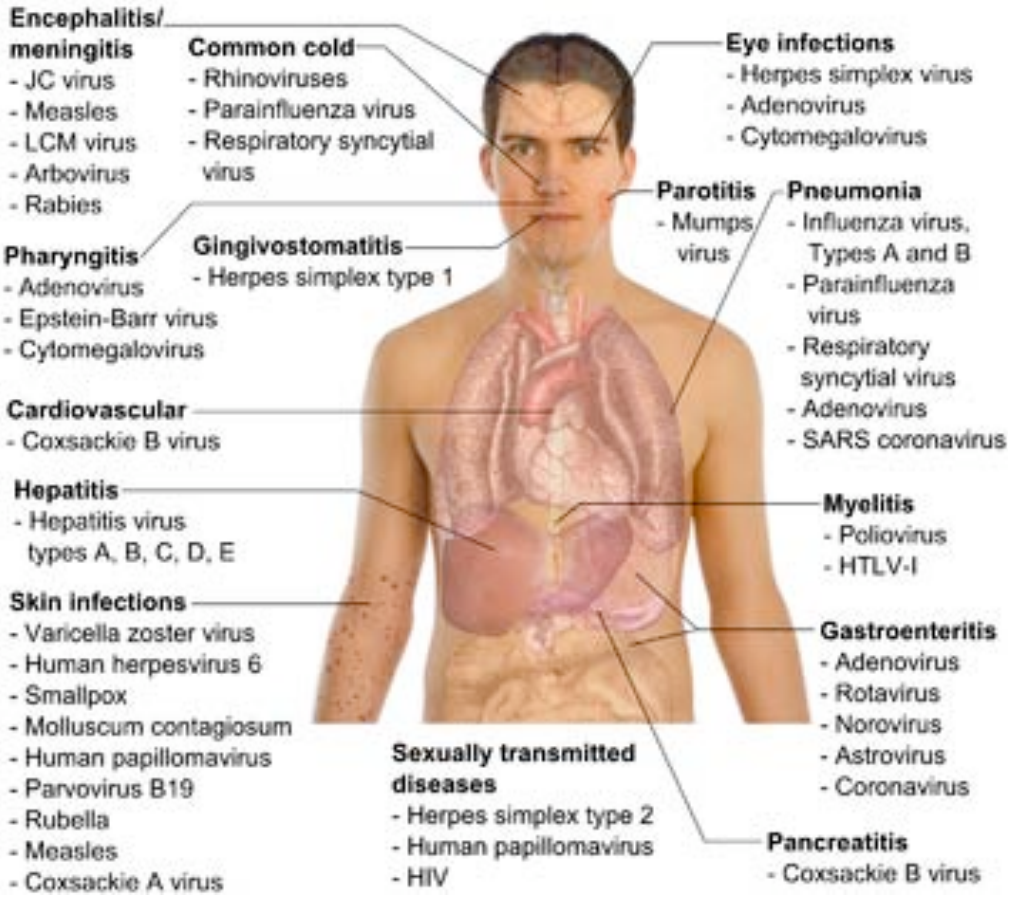
Viral Infections



Examples of Viral Targets

- Acute infections
  - Influenza
    - Timing issues - seasonal
  - Respiratory syncytial virus (RSV)
    - Patient population - pediatric, elderly
- Chronic infections
  - Human immunodeficiency virus (HIV)
    - Managed by long-term viral suppression with HAART
  - Hepatitis C (HCV)
    - Can “cure” with standard of care (pegylated interferon and ribavirin)
    - Not well tolerated, moderately efficacious (~50% in GT1)

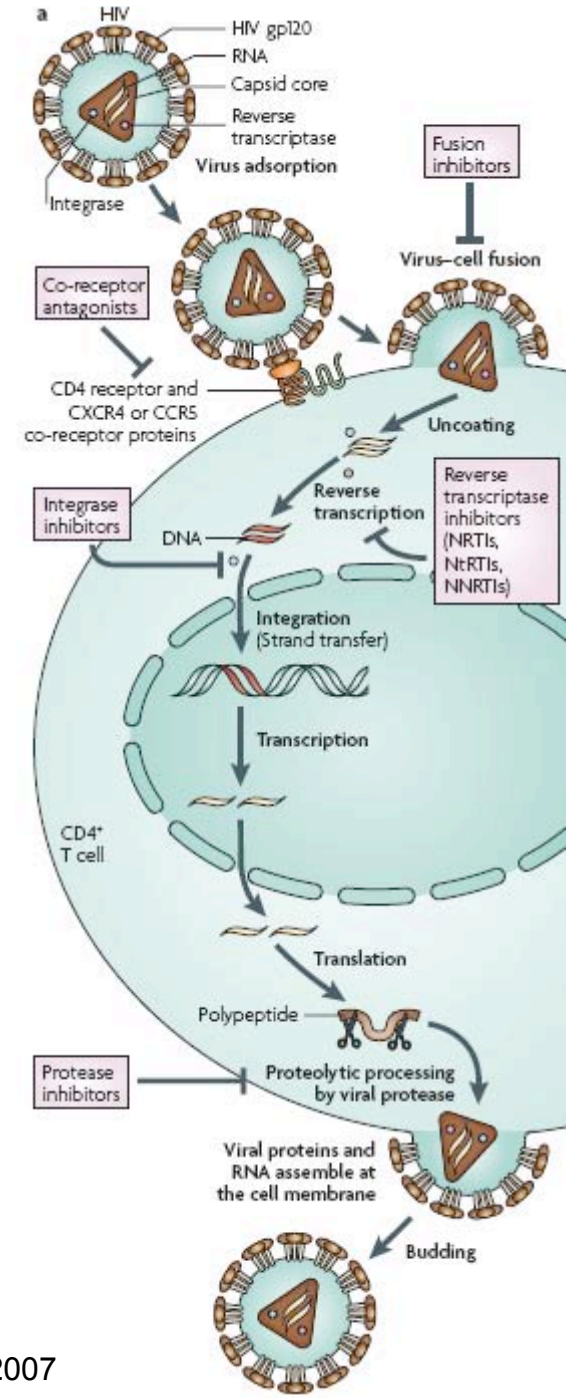
HIV

- World's leading infectious killer
- Each year, about 40,000 new HIV infections occur in the United States
- Globally, more than 33 million people are infected with HIV, including 2 million children
- In 2008, more than 2 million people died of AIDS-related causes. During the same year, 2.7 million new cases of HIV were reported.
- More than 25 million people have died of AIDS since the epidemic emerged in the early 1980s

Sources: Centers for Disease Control and Prevention, the World Health Organization

HIV therapeutic targets:

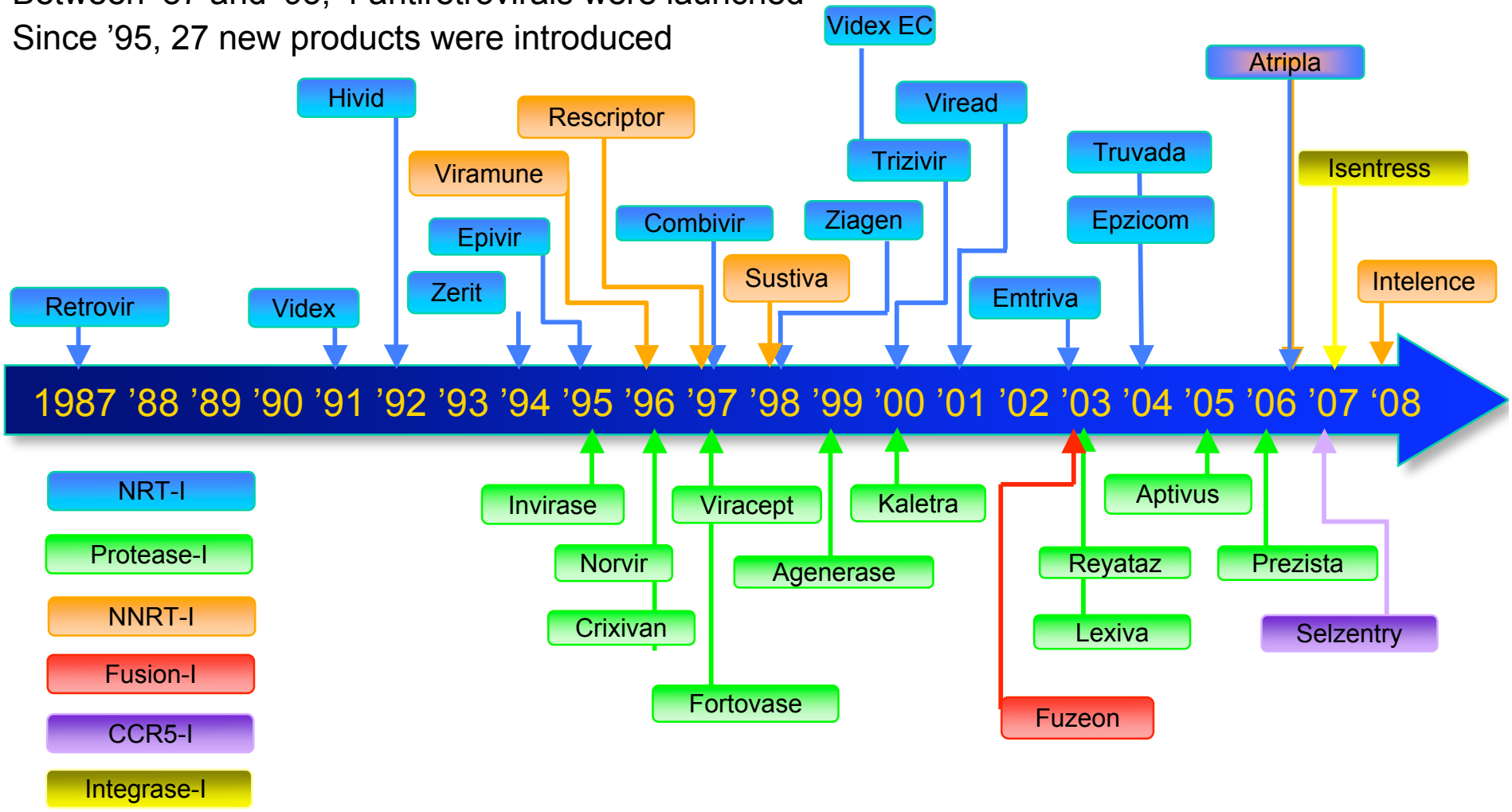
Multiple stages of the viral life cycle



From E. De Clercq, Nature Reviews, pp 1001-8, 2007

Approved Antiretrovirals for HIV

Between ’87 and ’95, 4 antiretrovirals were launched
Since ’95, 27 new products were introduced



http://www.fda.gov/oashi/aids/virals.html

HIV: Global Nonclinical Program

- Phase 1 POC trials; monotherapy for 7-14 days
  - Standard IND package (14/28 day studies, genotoxicity, safety pharmacology)
- Phase 2; 48-96 wks
  - Chronic studies needed early in development
  - Drug supply adequate
  - 28 day directly to chronic studies; 13 week interim to support clinical progression
  - Teratology for EU (See M3 (R2))
  - Consider immunotoxicity study (ICH S8)
  - Carcinogenicity range-finding/planning
  - Pediatric Investigation Plan (PIP for EU)
- Phase 3
  - Fertility
- Marketing application
  - Perinatal
  - Carcinogenicity
  - Environmental assessment

Carcinogenicity Studies

- Do carcinogenicity studies need to be completed prior to submitting an NDA/MAA?
  - US: Following ICH S1A
    - "for pharmaceuticals developed to treat certain serious diseases, carcinogenicity need not be conducted before market approval…"
  - EU: "Guideline on the carcinogenicity evaluation of the medicinal products for the treatment of HIV infection" (EMEA/CHMP/SWP/194898/2006)
    - Came in effect July 2008
    - Reconsiders the need and timing for carcinogenicity studies based on "nearly all" agents exhibiting positive findings in 2 yr carcinogenicity studies and the increased life expectancy of HIV-infected patients
    - Carcinogenicity to be submitted before granting the marketing authorization except for "products intended for the treatment of patients with limited treatment options or a clearly demonstrable added value"

Juvenile Toxicity Studies

- Are juvenile toxicity studies required?
- Guidance documents
  - "Nonclinical Safety Evaluation of Pediatric Drug Products" (FDA, Feb 2006)
  - "Guideline on the Need for Nonclinical Testing in Juvenile Animals of Pharmaceuticals for Pediatric Indications" (EMEA/CHMP/SWP/169215/2005, Aug 2008)
  - ICH M3 (R2)
- Guidelines are similar - Considerations include:
  - Tox package available, clinical data in adults
  - Pediatric population
  - Target organs relevant to developing systems
  - Case-by case decision on need for juvenile tox

Additional EU guidance

- Draft: “Reflection Paper on *In Vitro* Investigation of Mitochondrial Toxicity of Anti-HIV Nucleoside Reverse Transcriptase Inhibitors” (CHMP/SWP/8212/2007)
  - Recommends specific in vitro studies (cell lines, duration, endpoints)
- “Guideline on the Environmental Risk Assessment of Medicinal Products for Human Use” (EMEA/CHMP/SWP/4447/2000, Dec 2006)
- “Guideline on Detection of Early Signals of Drug-Induced Hepatotoxicity in Nonclinical Studies” (EU Draft EMEA/CHMP/SWP/150115/2006)

Nucleoside Reverse Transcriptase Inhibitors

- Act as alternate substrates for the RT and cause termination of the formation of the DNA chain
- Adverse effects associated with mitochondrial toxicity
  - Lactic acidosis, hepatic steatosis, neuropathy, myopathy, pancreatitis and lipodystrophy
  - Mechanisms: DNA polymerase γ inhibition, oxidative stress, mtDNA mutations
  - In vitro models
    - Kinetics of incorporation by DNA polymerase γ
    - Cell culture: e.g. HepG2, skeletal muscle, renal proximal tubule

Correlation of in vitro assays and clinical outcome

Compound	Inhibition of Pol _	Inhibition of mtDNA	Lipid content	Clinical Toxicities
zalcitabine	+++	++++	ND	No longer used
didanosine	+++	++	++	pancreatitis, peripheral nephropathy, lactic acidosis (hepatic steatosis)
stavudine	+++	+++	+++	peripheral nep hropathy, lipodystrophy, pancreatitis, lactic acidosis (hepatic steatosis), hyperlipidemia
zidovudine	+	-	+	bone marrow suppression (anemia), lactic acidosis (hepatic steatosis)
lamivudine	+	-	-	minimal toxicity
emtricitabine	+	-	-	minimal toxicity
tenofovir DF	+	-	-	renal insufficiency
abacavir	+	-	++	hypersensitivity

Venhoff et al, Antiviral Therapy, 2007;
Guidelines for the Use of Antiretroviral Agents in HIV-1-Infected Adults and Adolescents

NonNucleoside Reverse Transcriptase Inhibitors (NNRTIs)

- NNRTIs bind RT directly and inhibit action
- Toxicities of approved agents
  - Delavirdine: rash, incr. transaminases, headaches
  - Efavirenz: rash, incr. transaminases, CNS symptoms, teratogenic (monkeys)
  - Etravirine: rash, nausea
  - Neviripine: rash, symptomatic hepatitis/hepatic necrosis
- Preclinical models detected liver effects
- Rash, CNS effects not detected
  - Nevirapine model in female Brown Norway rats (Uetrecht et al , U of Toronto)

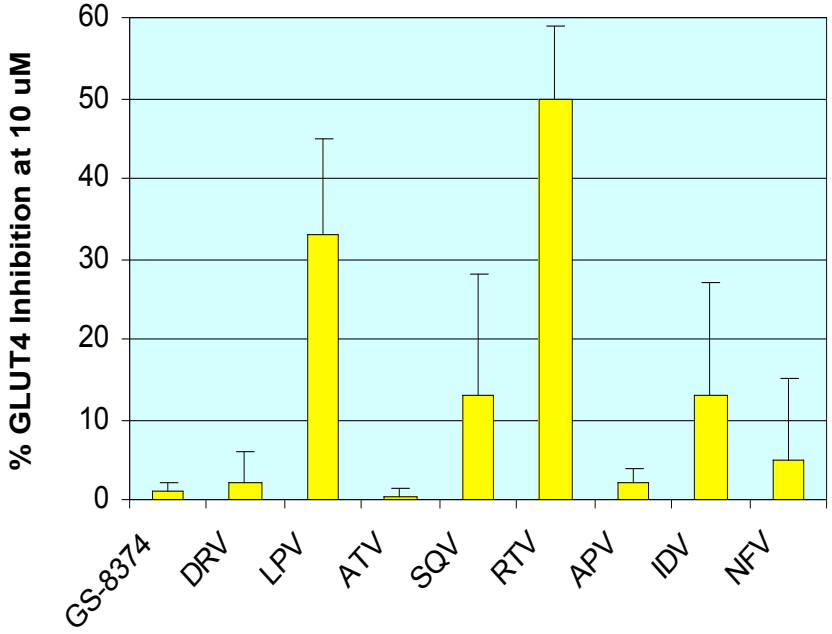
Protease Inhibitors (PIs)

- PIs inactivate the HIV-1 protease and prevent cleavage of gag-pol proteins
- Common 3rd Agent
  - atazanavir, darunavir, fosamprenavir, indinavir, lopinavir, nelfinavir, ritonavir, saquinavir, tipranavir
- Due to low exposure agents generally require “boosting” with ritonavir
- Side effect profiles include:
  - Hyperglycemia and fat maldistribution – all PIs
  - Hyperlipidemia – most PIs
  - GI intolerance – most PIs
  - Hepatotoxicity – darunavir, tipranavir
  - Skin rash – darunavir, fosamprenavir, tipranavir
  - Hyperbilirubinemia, prolonged PR - atazanavir

Screening for metabolic effects:
In Vitro Metabolic Toxicity in Adipocytes

- Differentiated human and mouse adipocytes

Insulin-stimulated glucose uptake
(Inhibition at 10 µM)



Lipid accumulation

PI	IC_{50} [µM]
GS-8374	>30
ATV	>30
DRV	>30
APV	>30
IDV	>30
RTV	17 ± 8
SQV	16 ± 4
LPV	16 ± 5
NFV	8 ± 3

Callebaut et al, CROI 2008

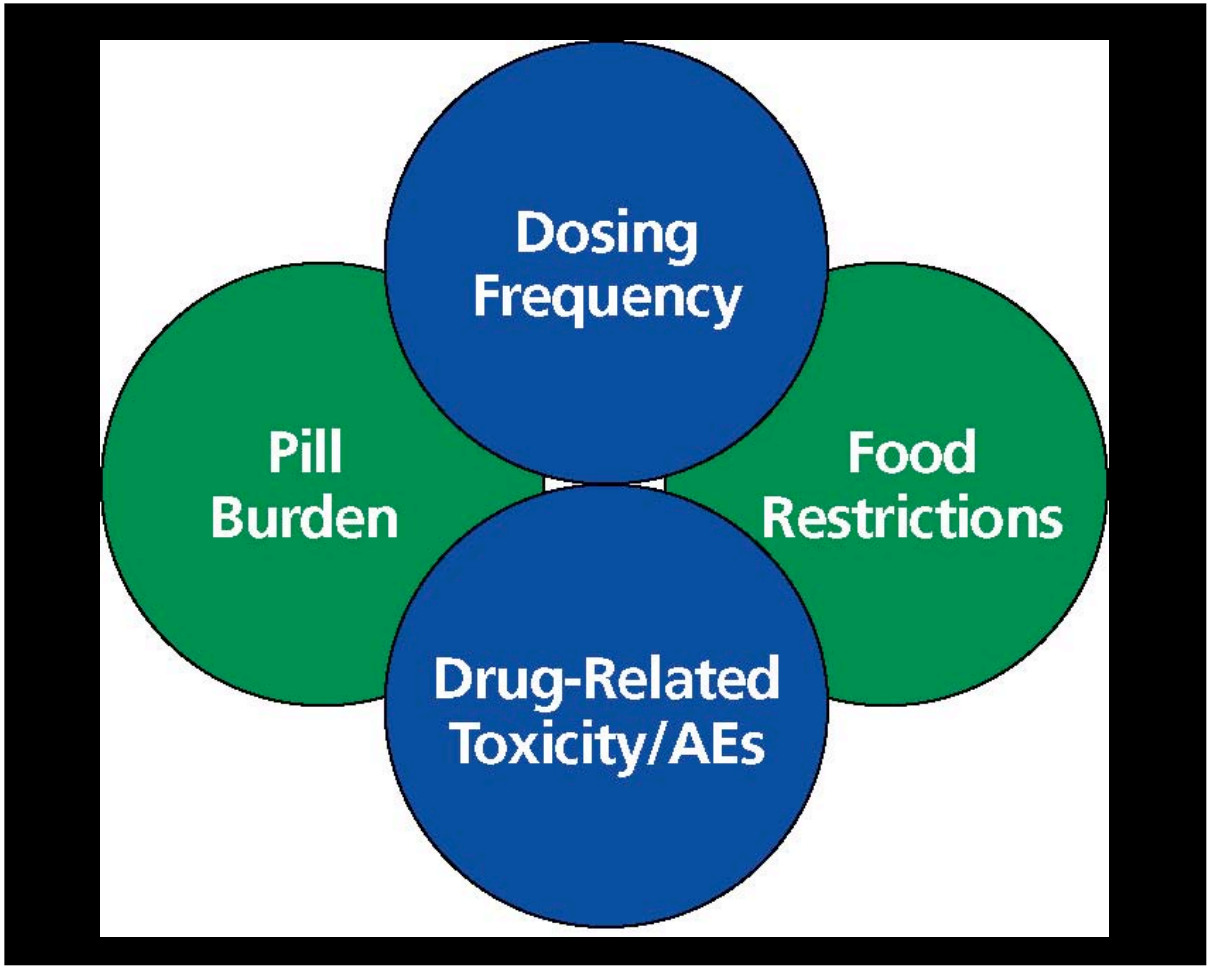
Other agents

- Fusion inhibitors
  - enfurvirtide: local injection site reactions (given sc), hypersensitivity
- CCR5 antagonists
  - maraviroc: abdominal pain, cough, dizziness, musculoskeletal symptoms, pyrexia, rash, upper respiratory tract infections, hepatotoxicity, orthostatic hypotension
- Integrase inhibitors
  - raltegravir: creatine kinase elevations
  - elvitegravir: being developed with a boosting agent

Combination Toxicity Studies

- HAART is based on multi-drug regimens
- Guidance documents:
  - US: “Nonclinical Safety Evaluation of Drug or Biologic combinations” (March 2006)
  - EU: “Guideline on the Nonclinical Development of Fixed Combinations of Medicinal Products” (EMEA/CHMP/SWP/258498/2005; in effect August 2008)
  - ICH M3 (R2)
- Late stage products: combination toxicity generally not needed
- Early stage products: varied guidance
  - Consider overlapping toxicities
  - Need for combination tox dependent on amount of nonclinical and clinical data with individual agents
  - 90 day combination tox in one species recommended in some cases
  - Other assessments, e.g. genotoxicity or embryo-fetal toxicity, dependent on characteristics of compounds and regional requirements

Regimen-Related Drivers of Adherence



DHHS. *Guidelines for the Use of Antiretroviral Agents in HIV-1 Infected Adults and Adolescents*.

Fixed Dose Combinations

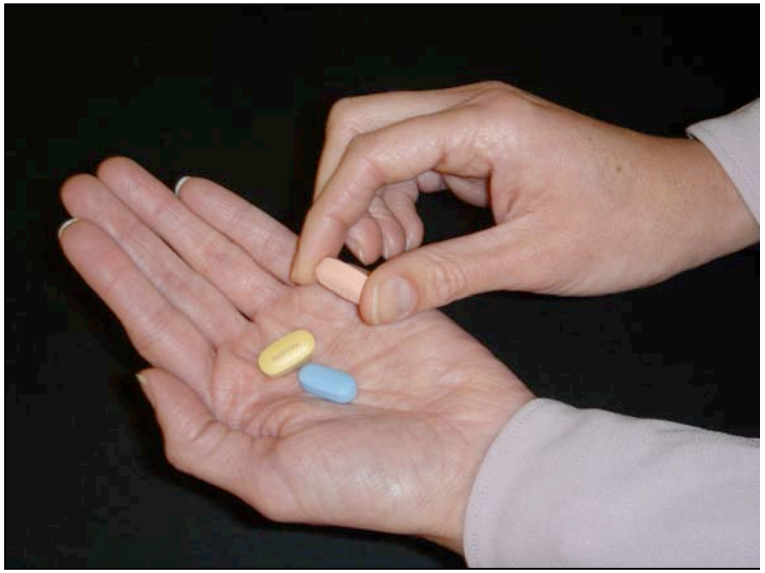
- Guidance documents:
  - "Fixed Dose Combinations, Co-Packaged Drug Products, and Single-Entity Versions of Previously Approved Antiretrovirals for the Treatment of HIV" (FDA Oct 2006)
    - Encouraged the development of FDC and delineated requirements
  - "Guideline on Clinical Development of Fixed Combination Medicinal Products" (CHMP/EWP/240/95 Rev.1 Sept 2009)
  - "Guideline on the Clinical Development of Medicinal Products for the Treatment of HIV Infection" (EMEA/CHMP/EWP/633/02 June 2009)

One Pill, Once a Day, Triple Combination Regimen

- December 20, 2004: Joint venture to develop and commercialize the fixed-dose combination of Sustiva® (efavirenz) and Truvada™ (tenofovir DF and emtricitabine) in the US
  - First multi-company effort to create fixed-dose product with three HIV/AIDS medicines
  - Multiple bioequivalence studies conducted
  - FDA approved on 7/12/2006 as the first complete HIV regimen available in once-daily fixed-dose combination

GILEAD

Bristol-Myers Squibb Company

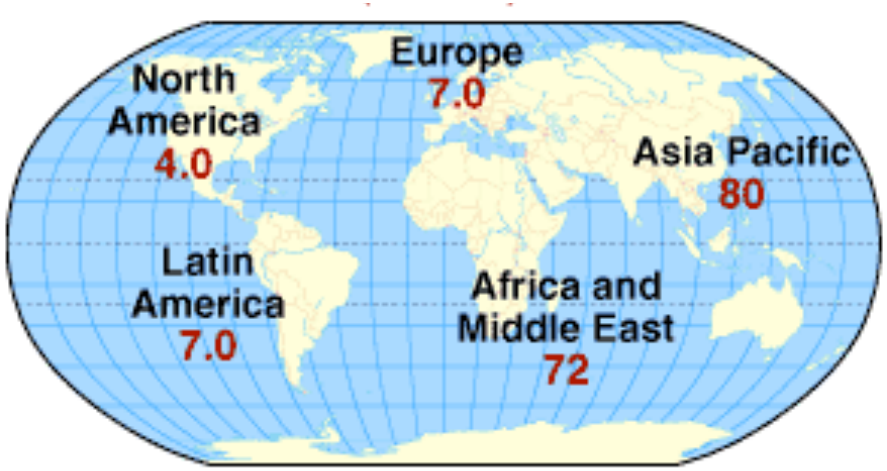


TDF + FTC + EFV = 1 Pill

HIV: The Future

- Treatment
  - Agents with long-term safety and improved resistance profiles still needed
  - Focus on regimen simplification
  - Nucleoside-sparing regimens
  - New targets
- Prevention
  - Vaccines
  - Pre-exposure prophylaxis (PrEP)
- HIV cure?

Chronic Hepatitis C:
A Significant Unmet Medical Need



Estimated Total Chronic HCV Infections Worldwide:

170 MILLION

- **More than 20% of chronic HCV infections lead to cirrhosis**
- **The leading cause of liver transplantation in the US**
- **HCV accounts for 30% of end-stage liver disease and liver cancer**
- **US: ~10,000 HCV-related deaths/year**

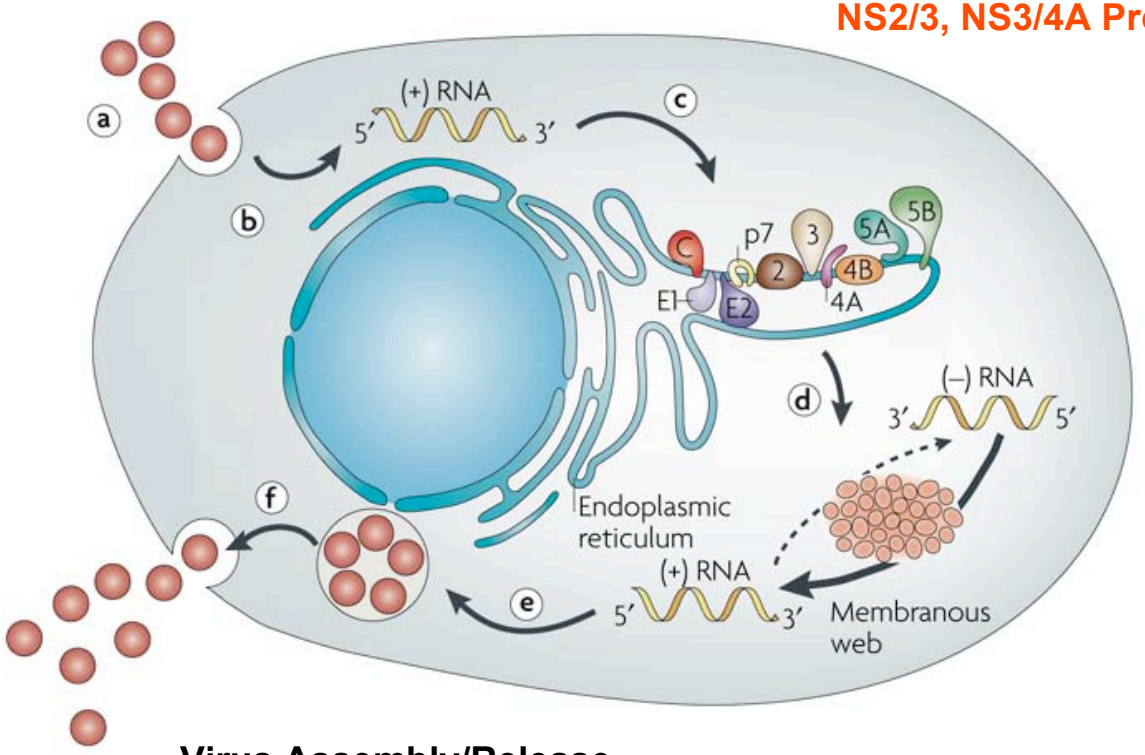
Standard of Care (SOC) associated with numerous toxicities

- Regimen is 24 (GT2/3) to 48 (GT1) weeks
- Pegylated Interferon
  - Flu-like syndrome (fatigue, fever, headache, myalgia)
  - Neuropsychiatric events
  - Bone marrow toxicities (↓ neutrophil, platelet counts)
  - Local injection site reactions
- Ribavirin
  - Hemolytic anemia
  - Teratogenic/embryocidal
  - Mutagenic

Drug Targets in the HCV Lifecycle:

HCV Binding/Entry
Entry Inhibitors

Translation/Polyprotein Processing
IRES Inhibitors
NS2/3, NS3/4A Protease Inhibitors



Replication Complex Formation/ RNA Replication
NS3 Helicase Inhibitors
NS4B Inhibitors
NS5A Inhibitors
NS5B Pol Nuc, Non-nuc, & Cyclophilin Inhibitors

Virus Assembly/Release
Glucosidase Inhibitors

Nature Reviews | Microbiology

Development of New HCV agents

- Regulatory path is not well established for direct acting antivirals (DAAs)
- Combination therapy will be required
  - Combination with PEG + RBV (SOC)?
  - Multiple DAAs?
- Considerations for early clinical trials
  - Limited monotherapy (rapid resistance)
  - Explore duration of therapy (differ from SOC?)
  - Which population to study
    - Naives, relapsers, partial/null responders, genotype, IL28B polymorphism?
  - Sustained virological response (SVR) data needed
- "Guideline on the clinical evaluation of direct acting antiviral agents intended for the treatment of chronic hepatitis C" EMEA/CHMP/EWP/30039/2008

HCV: Global Nonclinical Program

- Phase 1 POC trials; monotherapy for 3-5 days
  - Standard IND package (14/28 day studies, genotoxicity, safety pharmacology)
- Phase 2; 4/12/24 weeks
  - Duration varies with goals of Phase 2
  - Repeat dose tox to match trial design or support combination dosing
  - Combination tox considerations
  - Teratology for EU (See M3 (R2))
  - Carcinogenicity range-finding/planning
  - Pediatric Investigation Plan (PIP for EU)
- Phase 3
  - Fertility
- Marketing application
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  - Carcinogenicity
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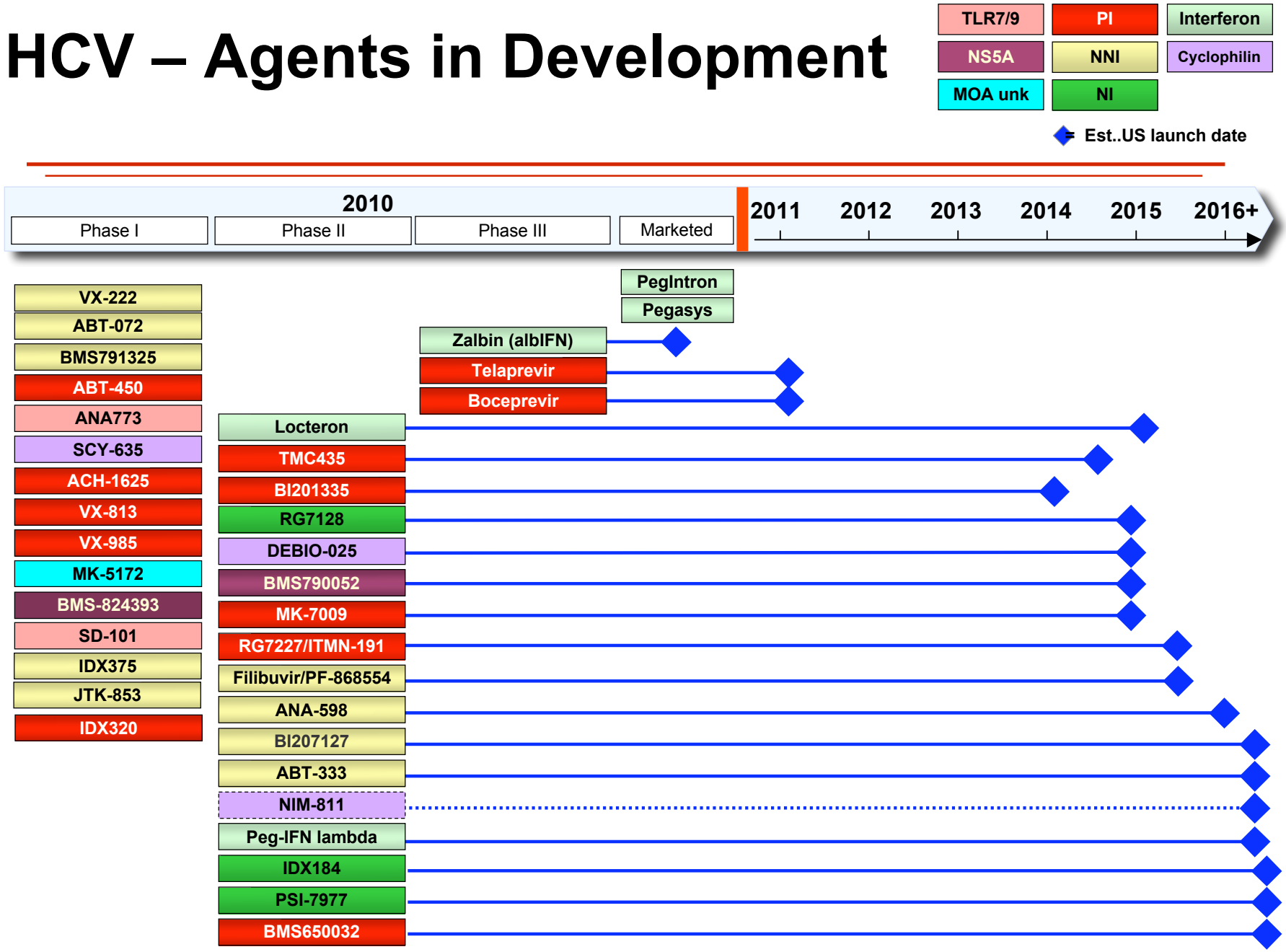
Many Unknowns for Combination DAAs

- Clinical studies with or without PEG/RBV?
- How many agents are needed to suppress virus and produce “cure” (SVR)?
- What nonclinical combination studies are required to support combination trials?
  - US: Nonclinical combination studies not useful; 3 month repeat dose studies with individual agents needed. Combination studies with PEG/RBV not needed.
  - EU: Some countries following ICH M3(R2) which requires combination studies
  - Evaluate overlapping tox, consider trial stage and design, make an argument based on data

HCV – Agents in Development



• For launch dates: assumption for competitor timelines is 2 yrs for phase 2b and 2.75 yrs from start of phase 3 to NDA submission

HCV: The Future

- New DAAs and new combinations
- Better response rates (SVR> 60%) and fewer relapses
- Shortened duration of treatment
- Reduced toxicities