Advanced HIV Resistance

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Disclaimer

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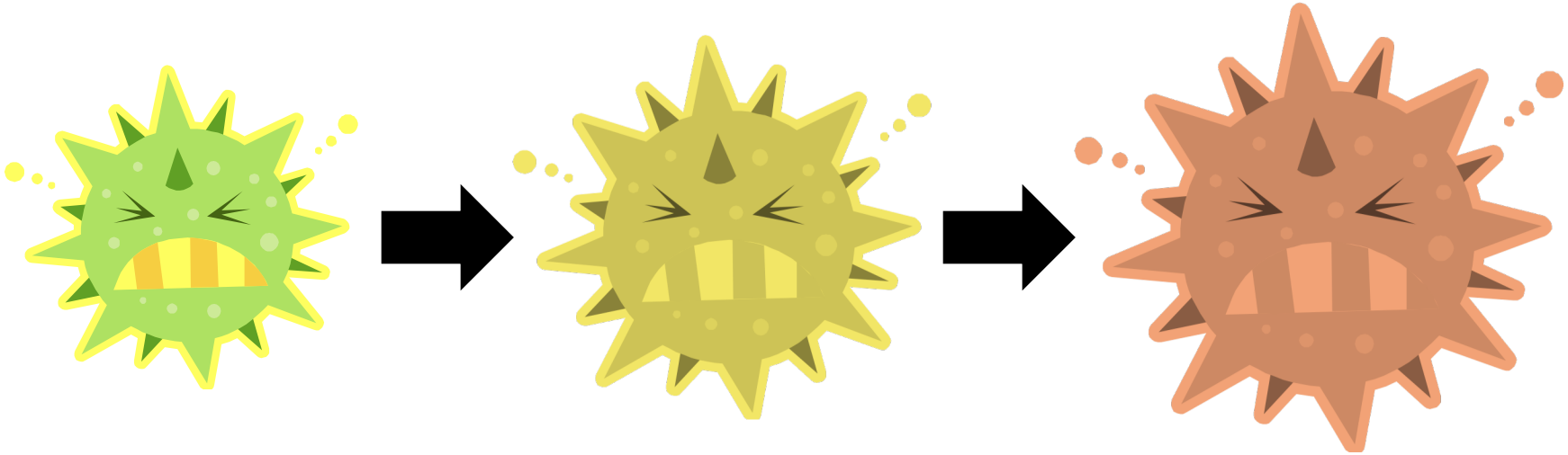
Outline

- Basic Resistance Review
- Types of resistance testing
- Review of specific HIV mutations
- Patient Cases

What Is Resistance?

- In short:
  - The HIV virus changes and the medications stop working
  - The more mutations present, the fewer medications are left available



How Do Mutations Develop?

High levels of viral production and turnover
10 million to 1 billion copies daily

Reverse transcription of viral RNA into DNA is highly error prone
Average 1 error in every genome

Complex mixture of viral quasispecies each with slightly different mutations

Clavel F et al. N Engl J Med 2004;350:1023-35. Markowitz M et al. J Virol 2003;77:5037-8. Roberts JD et al. Science 1988;242:1171-3.

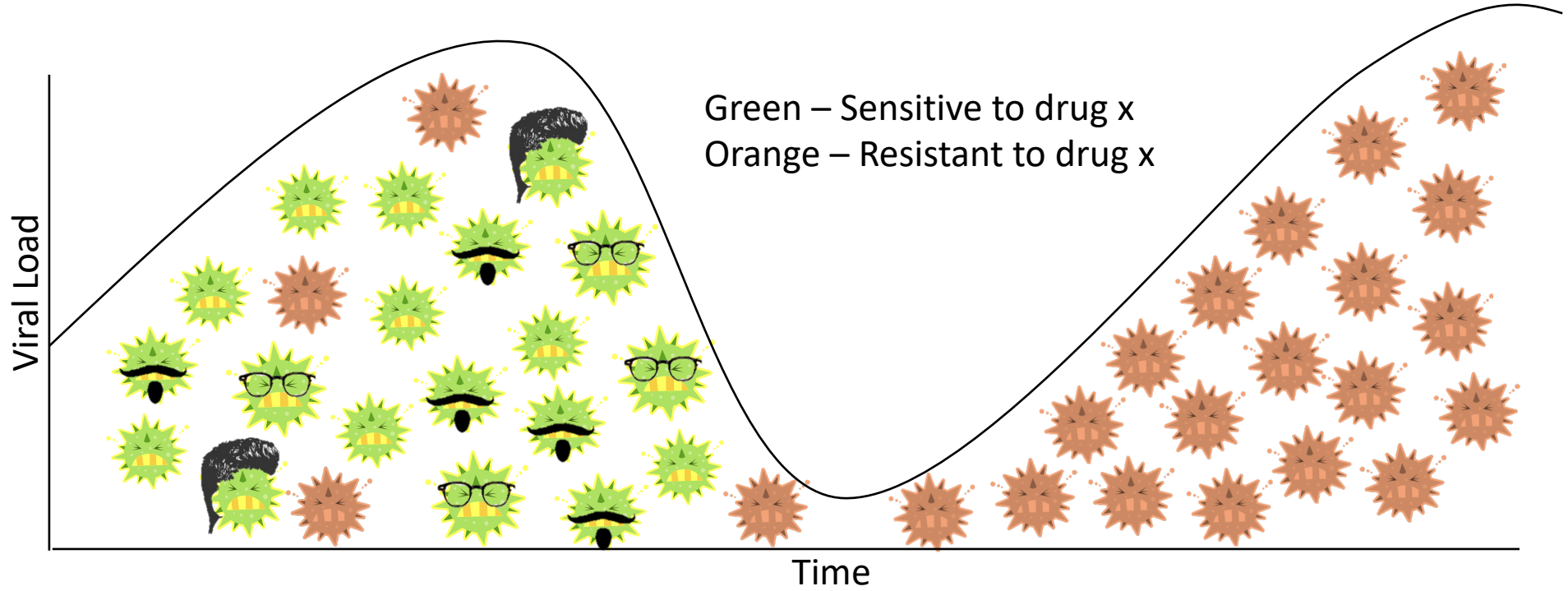
How Does Drug Resistance Occur?

- Option 1 – Develops in a patient taking antiretrovirals
- Option 2 – Transmitted during initial infection or subsequent reinfection

Clavel F et al. N Engl J Med 2004;350:1023-35.

Option 1 – Developed Resistance

- Resistant virus emerges when medication concentrations are:
  - Insufficient to suppress viral replication
  - Sufficient to cause a positive selective pressure



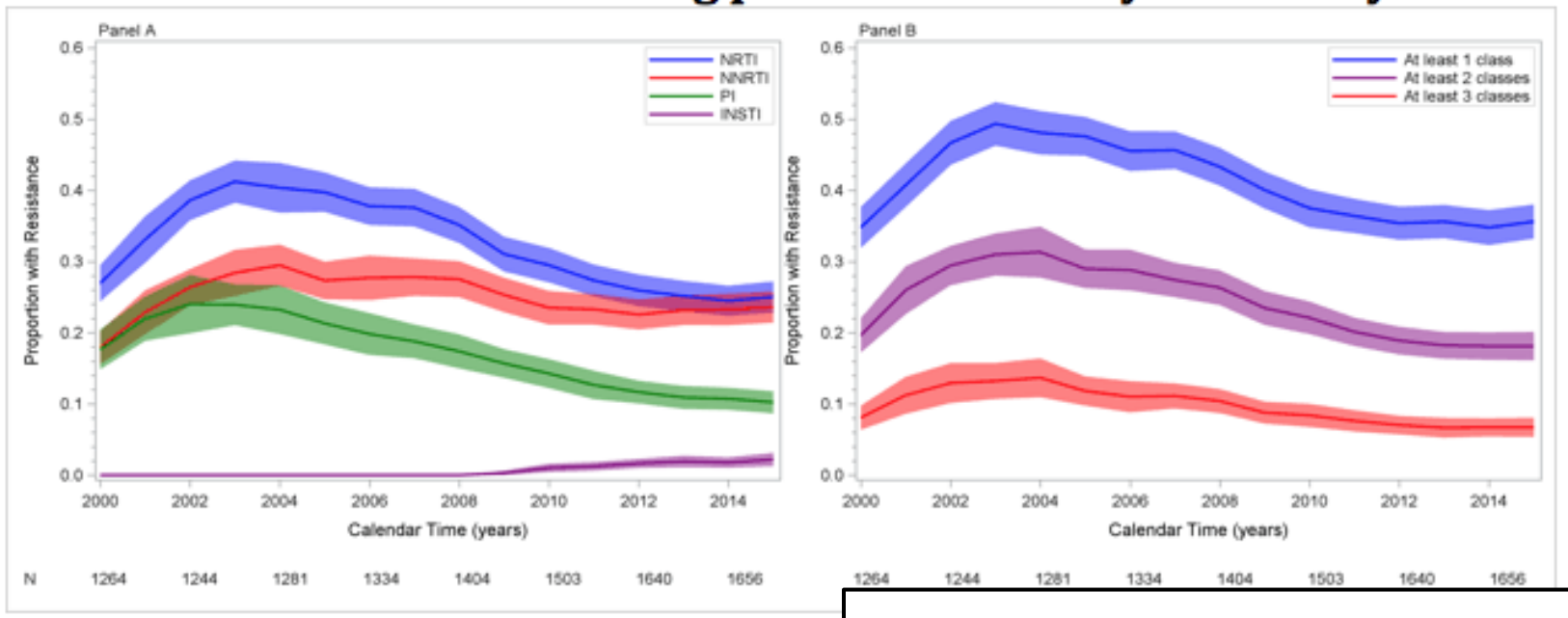
Cause of Developed Resistance

- Low concentrations of antiretrovirals in the body due to:
  - Non-adherence
    - Missed doses
    - Delayed doses
  - Inadequate absorption
  - Drug interactions

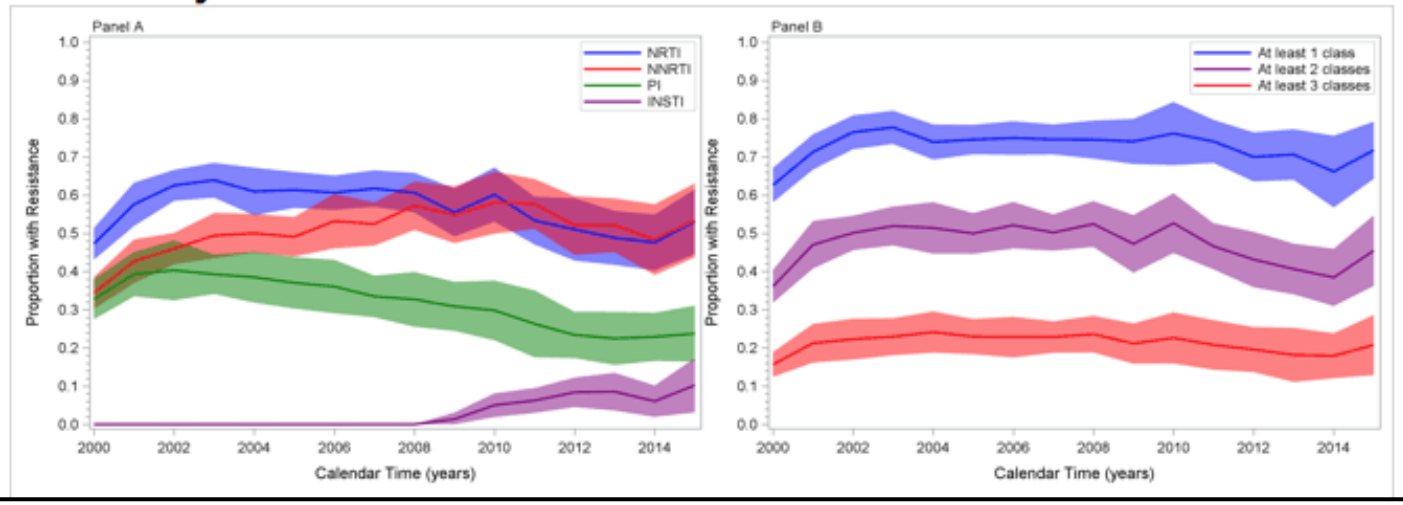
Option 2 – Transmitted Resistance

- Viral strains with drug resistance can be transmitted during initial infection or with subsequent re-exposure
- Encourage safe sex practices or clean needle use even after infected
- Patients fully adherent to therapy can develop resistance through exposure from a non-adherent partner

Prevalence of resistance among patients in care by calendar year.



Prevalence of resistance among patients with virologic failure by calendar year.



Davy T, Brunet L, Napravnik S, Zakharova O et al. Prevalence Of HIV Drug Resistance With Modern Agents. Conference on Retroviruses and Opportunistic Infections (CROI), February 13-16, 2017, Seattle. Abstract 483.

When to perform resistance testing

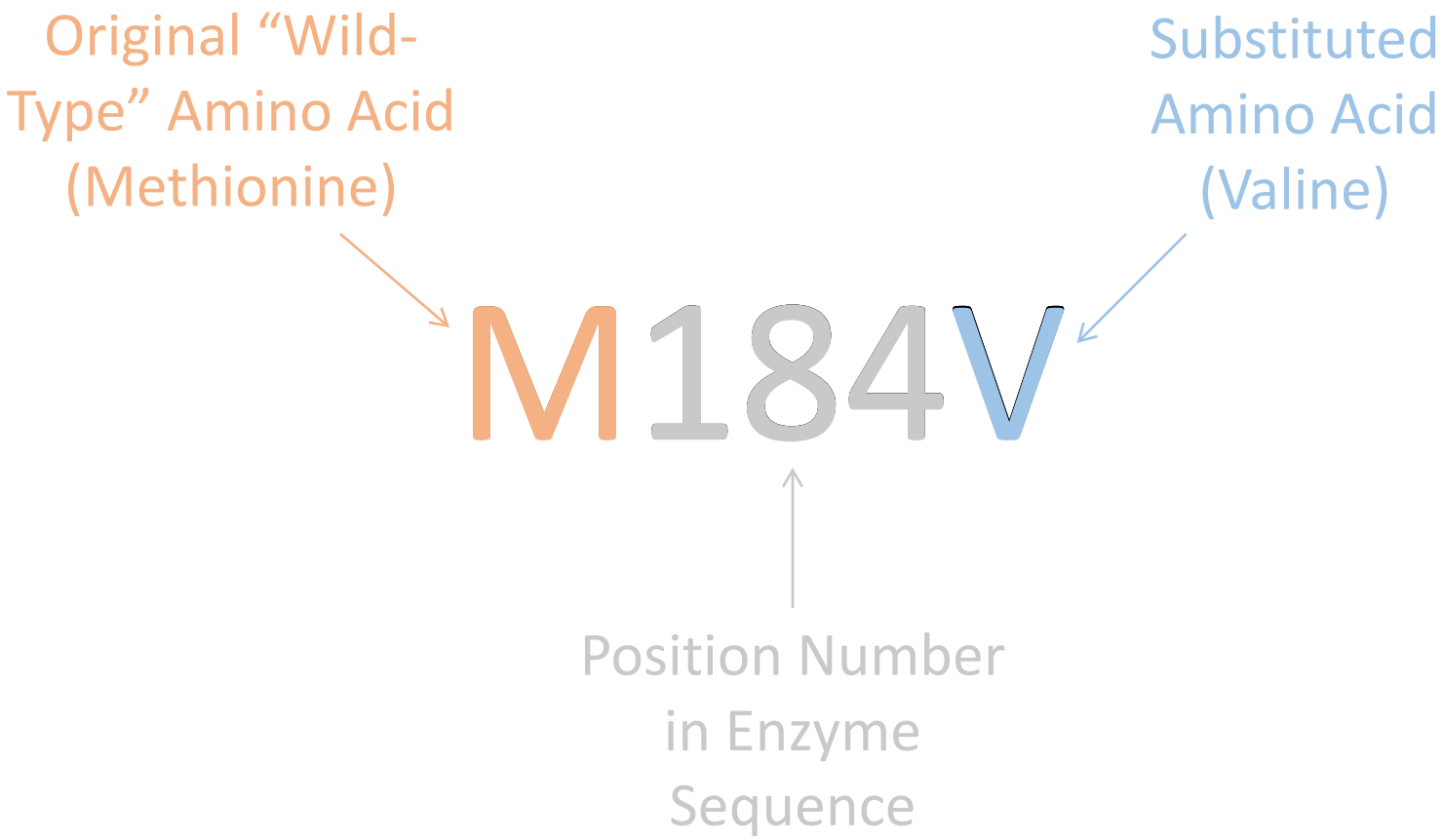
Clinical Setting	Rational/Comment
Acute HIV infection	• To determine if resistant virus was transmitted; guide treatment decisions • Consider repeat testing if treatment is deferred
In ART-naive patients with chronic HIV infection	• Transmitted drug-resistant virus is common in some areas; is more likely to be detected earlier in the course of HIV infection • Consider repeat testing at the time of starting ART
In patients with virologic failure	• To assist in selecting active drugs for a new regimen.
In patients with suboptimal suppression of viral load once on ART	• To assist in finding previously unknown mutations and guide future treatment decisions
Pregnancy	• Goal to achieve maximal viral suppression for prevention of perinatal transmission of HIV

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

HIV Genotype

- Sequences specific HIV enzymes
  - Reverse transcriptase, protease, integrase
- Detects mutations in specific codons that confer resistance
- Also detects naturally occurring mutations that do not confer resistance
- Computerized algorithms interpret test and determine antiretroviral activity

Mutation Terminology



Example Genotype

Last Name	Patient	**First Name**	Unknown
Clinic	UNKNOWN	**Physician**	Unknown
MR Number	123456	**Accession Number**	123456 (49138)
Collection Date	07/17/2010	**Received Date**	07/17/2010
Date Entered	07/17/2010	**File Name**	

Sequence includes PR codons: 1 - 99
Sequence includes RT codons: 1 - 299
There are no insertions or deletions
Subtype: B
No. previous patient sequences: **PR:** 0 **RT:** 0

PI Major Resistance Mutations	V82AV
PI Minor Resistance Mutations	None
Other Mutations	I64V, I72M

Protease Inhibitors

atazanavir/r (ATV/r)	Low-level resistance
darunavir/r (DRV/r)	Susceptible
fosamprenavir/r (FPV/r)	Potential low-level resistance
indinavir/r (IDV/r)	Intermediate resistance
lopinavir/r (LPV/r)	Low-level resistance
nelfinavir (NFV)	Intermediate resistance
saquinavir/r (SQV/r)	Potential low-level resistance
tipranavir/r (TPV/r)	Susceptible

NRTI Resistance Mutations	D67N, K70R, M184IMV, K219Q
NNRTI Resistance Mutations	V90IV, K103N, K238N
Other Mutations	V60I, K102R, D123E, T139KR, S162NS, K166KR, I178M, G196E, D237DN, A272S, T286A, E297K

Nucleoside RTI

lamivudine (3TC)	High-level resistance
abacavir (ABC)	Low-level resistance
zidovudine (AZT)	Intermediate resistance
stavudine (D4T)	Intermediate resistance
didanosine (DDI)	Potential low-level resistance
emtricitabine (FTC)	High-level resistance
tenofovir (TDF)	Potential low-level resistance

Non-Nucleoside RTI

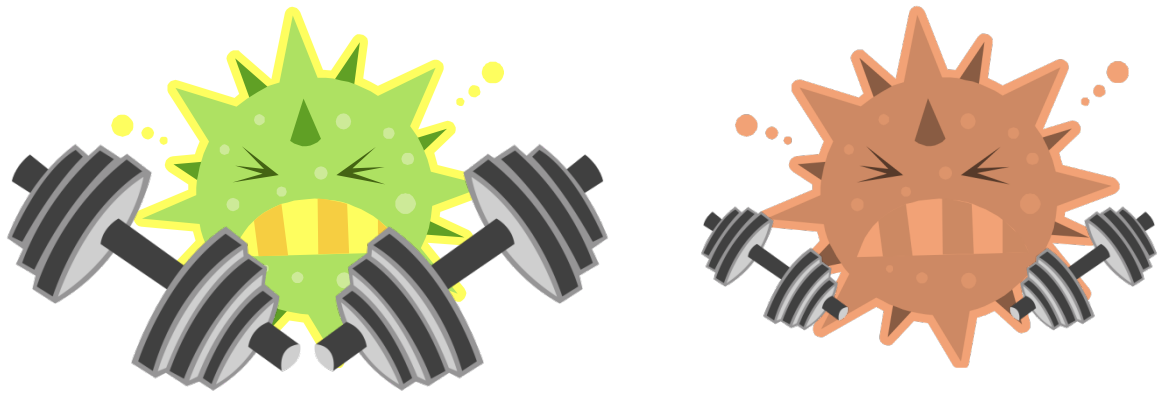
delavirdine (DLV)	High-level resistance
efavirenz (EFV)	High-level resistance
etravirine (ETR)	Potential low-level resistance
nevirapine (NVP)	High-level resistance

Genotype Pro and Cons

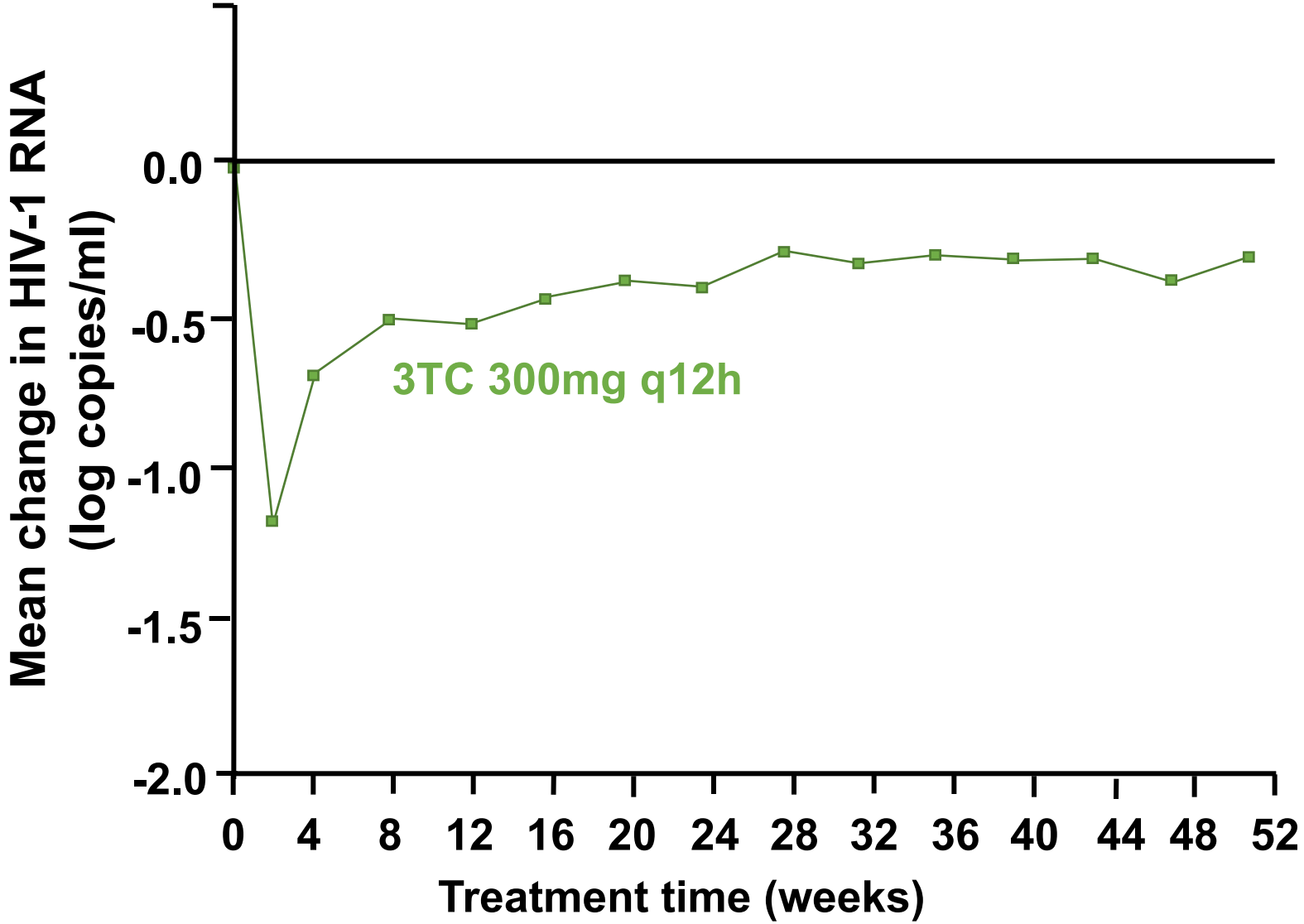
- Advantages
  - Cheaper
  - Results usually in 2-4 weeks
  - Good data on known mutations
- Disadvantages
  - Can be difficult to interpret when many mutations are present
  - Requires patient viral load to be >500 – 1000copies/mL
  - May miss archived mutations
  - Some tests may not update algorithms as fast

Viral Fitness

- Mutation-free, wild-type virus is the most “fit”
  - Virus replicates the fastest
- Mutations that confer drug resistance can decrease fitness
- Drug presence no longer inhibits replication, but maintaining mutations slows replication speed



Advantages of M184V mutation



Eron JJ, et al. NEJM, 1995; 333:1662-1669.

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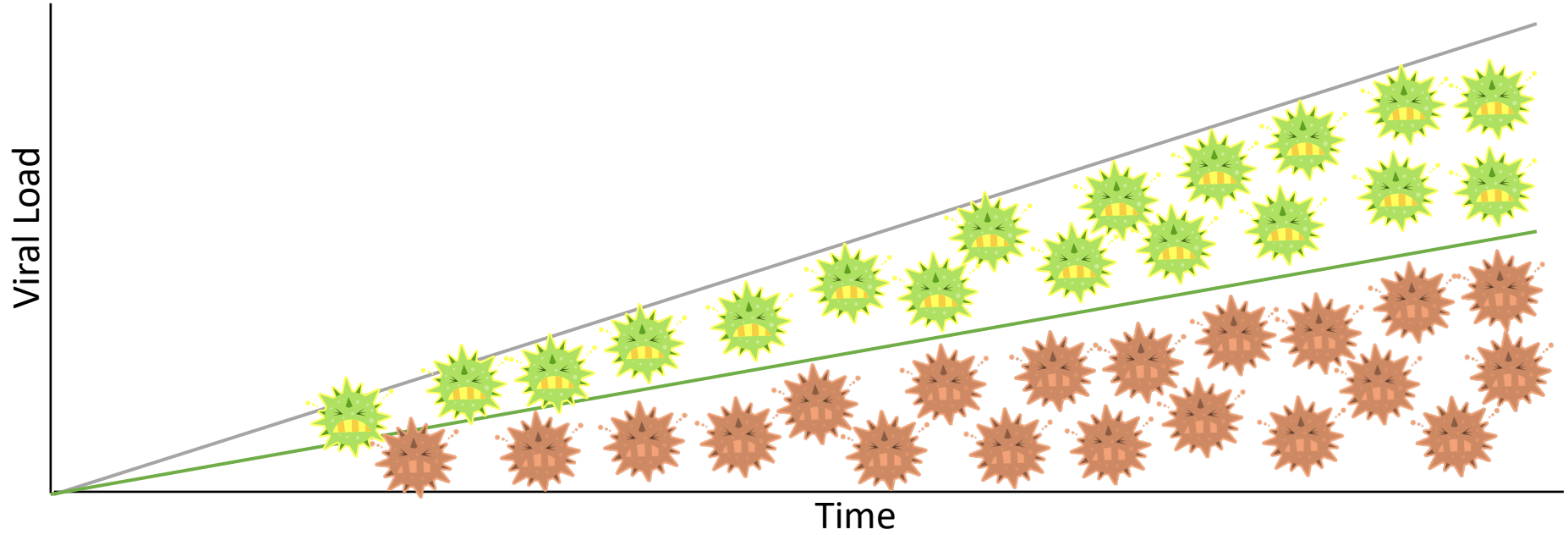
Archived Mutations

- Mutations developed by a patient that are not detected by a resistance test
- When drug pressure is removed, wild-type virus return as the dominant strand due to stronger fitness
- Mutation returns when drug pressure is resumed



Archive DNA Genotyping

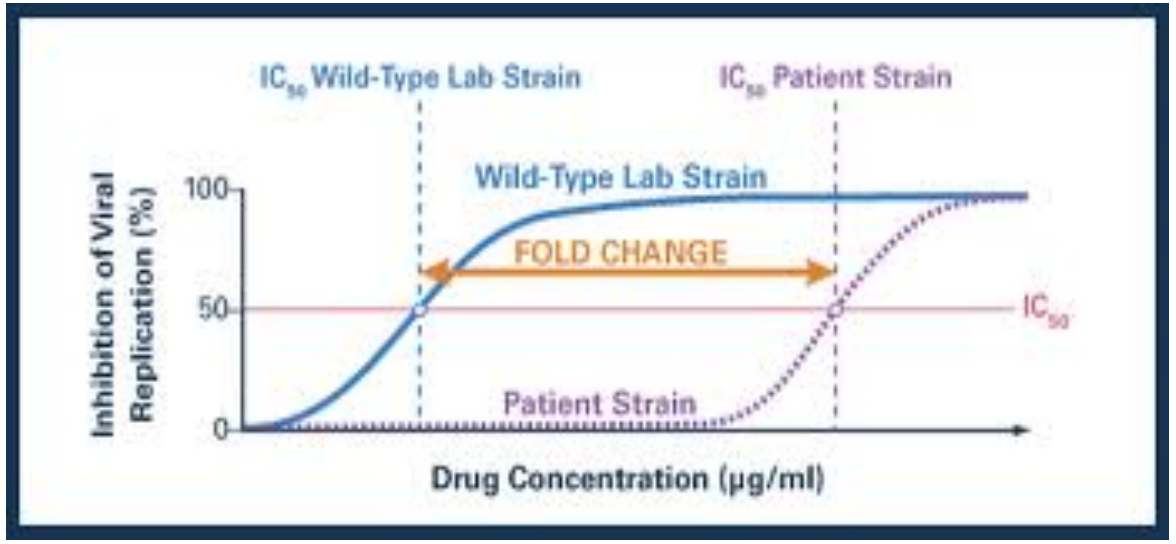
- Sequence the viral DNA either integrated or unintegrated in the cytoplasm
- Provide interpretations of drug resistance based on observed mutations similar to a standard genotype

Archive Genotype Pros and Cons

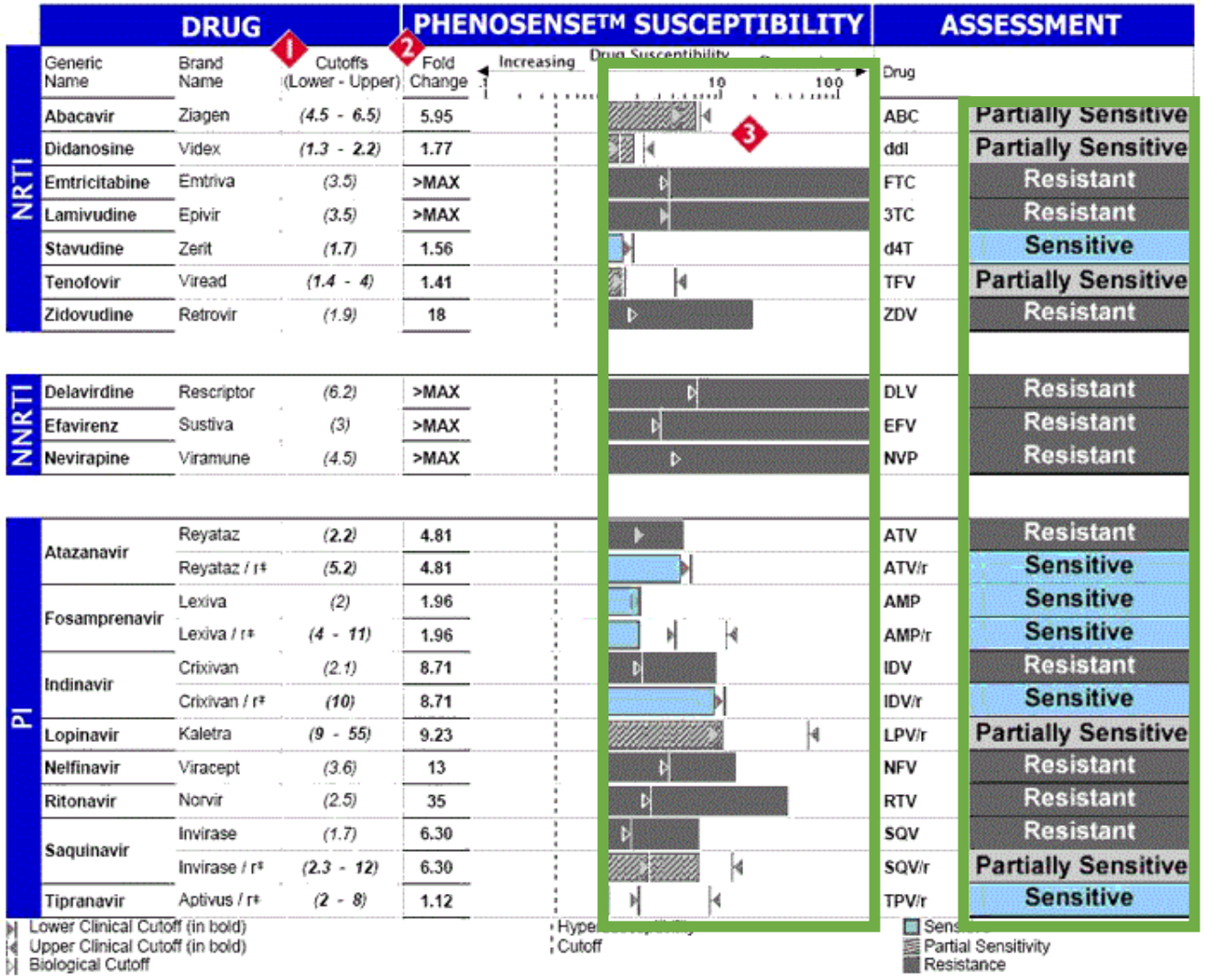
- Advantages
  - Can detect minority strains of the virus or “archived” mutations
  - Can detect mutations in patients even with a viral load <500
- Disadvantages
  - The clinical relevance of detecting minority strains is still uncertain
  - Can fail to detect already known past mutations observed on standard genotypes
  - Expensive

HIV Phenotype

- Measure ability of virus to grow in different drug concentrations
- Results expressed as “fold change” when compared to fully susceptible virus
- Higher resistance = higher drug concentrations needed to inhibit replication



Example of HIV Phenotype results



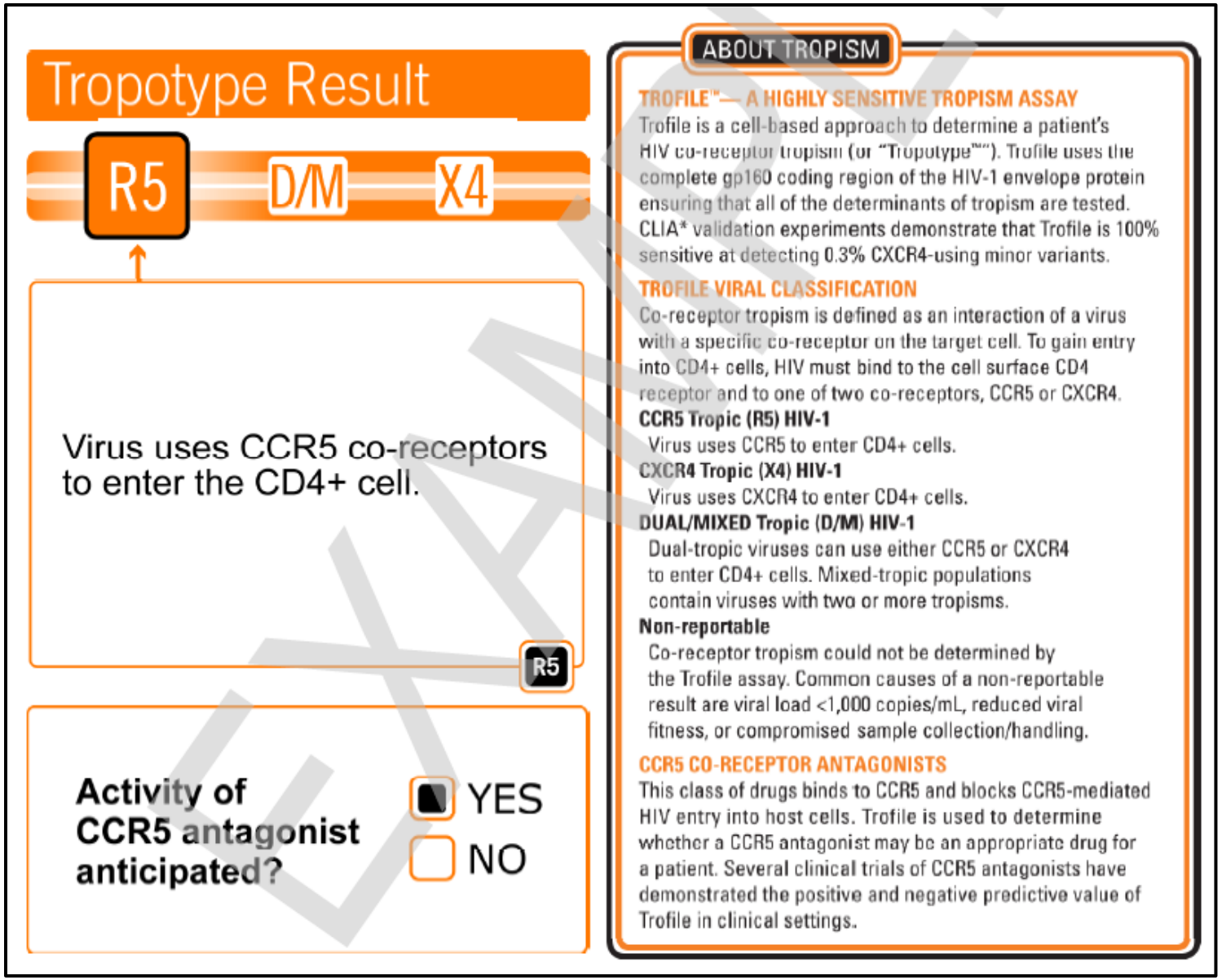
Phenotype Pros and Cons

- Advantages
  - Easy to read – need less knowledge of specific mutations
  - Provides quantitative information on resistance regardless of how many mutations present
- Disadvantages
  - More expensive
  - Requires patient viral load to be >500 – 1000
  - Clinically significant cut-offs for fold change have not been clearly defined for all drugs

HIV Trophile

- Used to determine if virus uses CCR5 or CXCR4 co-receptors (or both) to enter the cell
- Currently only used for maraviroc
- Expensive

HIV Trophile



Important Resistance Mutations

	Discriminatory Mutations					Thymidine Analog Mutations (TAMs)						MDR Mutations	
	184	65	70	74	115	41	67	70	210	215	219	69	151
Consensus	M	K	K	L	Y	M	D	K	T	T	K	T	Q
3TC	VI	R										Ins	M
FTC	VI	R										Ins	M
ABC	VI	R	E	VI	F	L			W	FY		Ins	M
DDI	VI	R	E	VI		L			W	FY		Ins	M
TDF	***	R	E		F	L		R	W	FY		Ins	M
D4T	***	R	E			L	N	R	W	FY	QE	Ins	M
ZDV	***	***	*	*		L	N	R	W	FY	QE	Ins	M

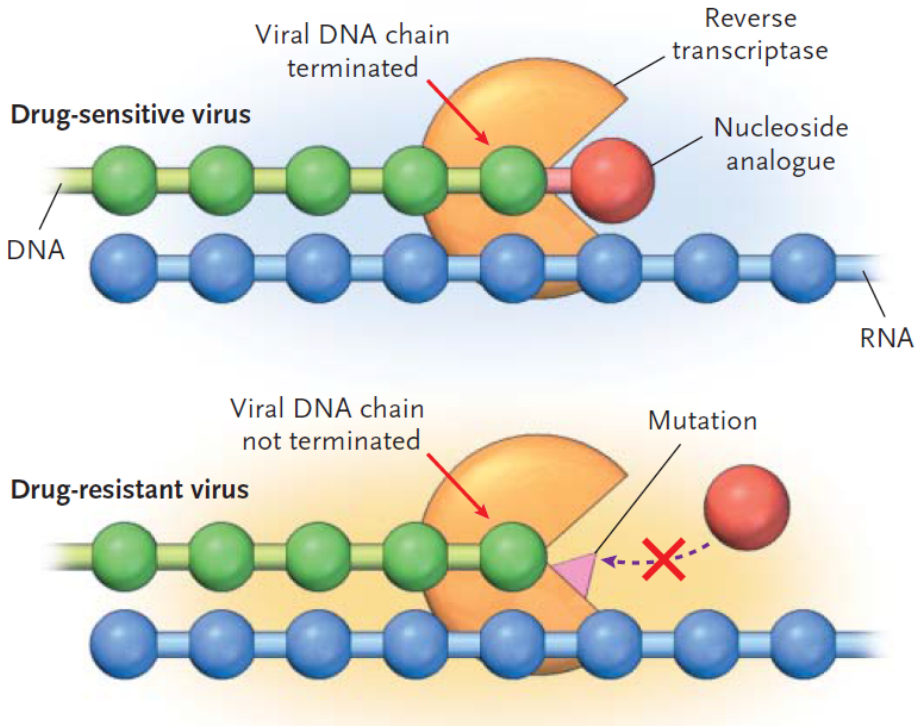
	100	101	103	106	138	181	188	190	230
Cons	L	K	K	V	E	Y	Y	G	M
NVP	I	EP	NS	AM		CIV	LCH	ASE	L
EFV	I	EP	NS	AM		CIV	LCH	ASE	L
ETR	I	EP			AGKQ	CIV	L	ASE	L
RPV	I	EP			AGKQ	CIV	L	ASE	L

	30	32	33	46	47	48	50	54	76	82	84	88	90
Cons	D	V	L	M	I	G	I	I	L	V	I	N	L
ATV/r		I	F	IL	V	VM	L	VTALM		ATFS	V	S	M
DRV/r		I	F		VA		V	LM	V	F	V		
FPV/r		I	F	IL	VA		V	VTALM	V	ATSF	V		M
IDV/r		I		IL	V			VTALM	V	AFTS	V	S	M
LPV/r		I	F	IL	VA	VM	V	VTALM	V	AFTS	V		M
NFV	N		F	IL	V	VM		VTALM		AFTS	V	DS	M
SQV/r						VM		VTALM		AT	V	S	M
TPV/r		I	F	IL	VA			VAM		TL	V		

	66	92	138	140	143	147	148	155
Consensus	T	E	E	G	Y	S	Q	N
Raltegravir (RAL)	A	Q	KA	SA	RCH		HRK	H
Elvitegravir (EVG)	IAK	Q	KA	SA		G	HRK	H
Dolutegravir (DTG)		Q	KA	SA			HRK	

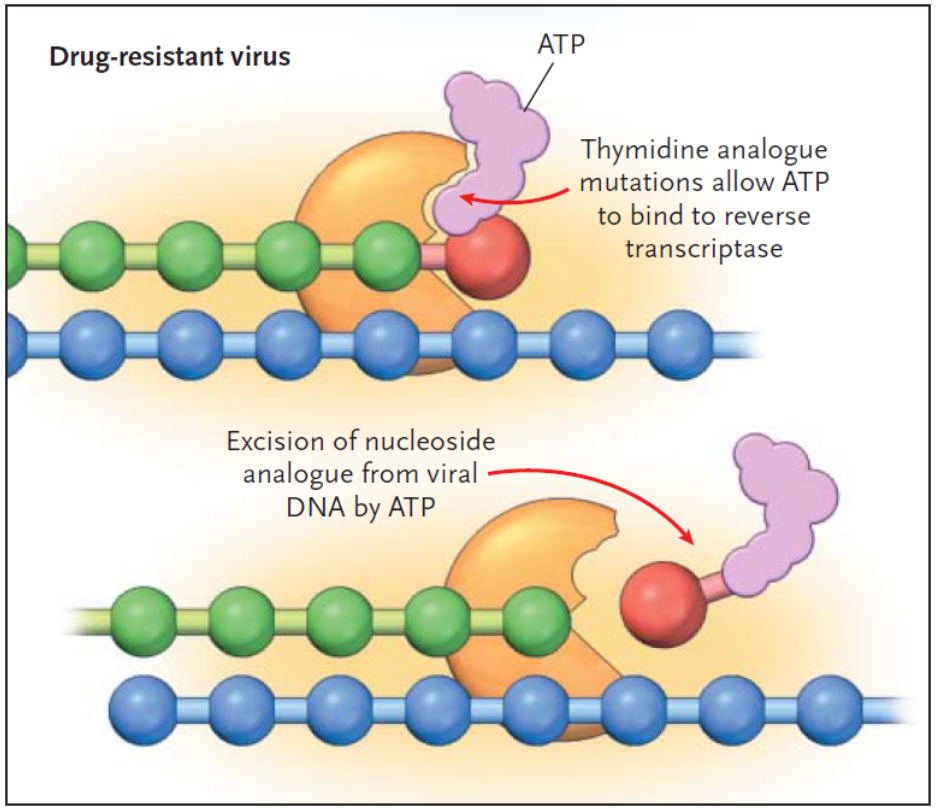
NRTI Mutations

- M184V
  - Selected by lamivudine and emtricitabine
    - >100 fold resistance to these medications
  - Low level resistance to abacavir and didanosine
  - Increases susceptibility to tenofovir, stavudine and zidovudine
  - Often maintained due to impact on viral replication



NRTI Mutations (cont.)

- Thymidine Analog Mutations – TAMs
  - Selected by zidovudine and stavudine
  - Reduces susceptibility to all NRTIs based on type and number of TAMs
  - Occur in 2 pathways:
    - M41L, L210W, and T215Y
    - D67N, K70R, T215F, and K219Q/E



NRTI Mutations (cont.)

- K65R/N/E
  - Selected by tenofovir, abacavir, didanosine and stavudine
  - In combination with M184V, can cause high level resistance to all NRTIs save for zidovudine
- T69i
  - Often occurs with multiple TAMs
  - Causes high level resistance to all NRTIs

NNRTI Mutations

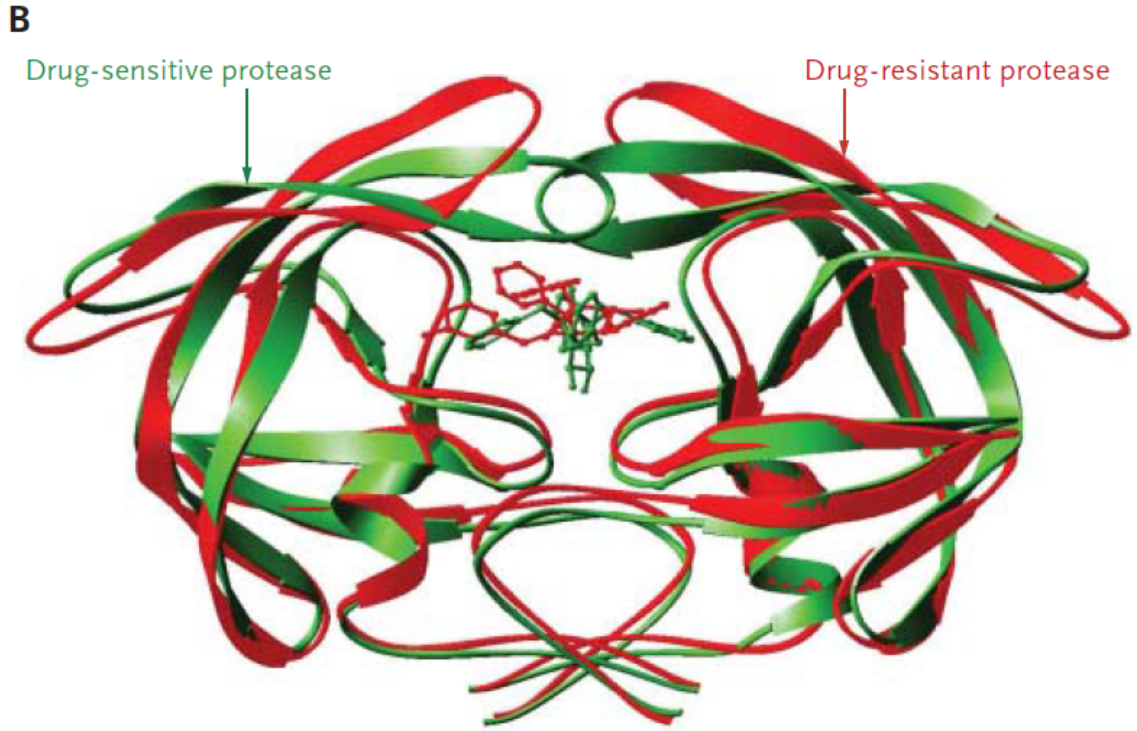
- K103N
  - Reduces nevirapine and efavirenz susceptibility by about 50 and 20-fold respectively
  - Easily obtained with non-adherence
  - No impact on etravirine or rilpivirine
- Y181C
  - Can be selected by and confer resistance to all NNRTIs save for doravirine to varying degrees

PI Mutations

- Higher barrier to resistance than other classes
- Resistance often requires multiple mutations
- Cross resistance is common
- Mutations are either major or minor
  - Major/primary – Confer drug resistance
  - Minor/accessory – attempt to recover lost viral fitness due to major mutations

PI Mutations (cont.)

- Enzyme active site “stretches” to decrease PI binding
- Medications with a higher barrier to resistance (darunavir) can “flex” to maintain activity



INSTI Mutations

Primary Mutation → **Accessory Mutations**

Q148R
- RAL – High Resistance
- EVG – High Resistance
- DTG – Low Resistance
- BIC – Low Resistance

→ **Q148R + G140A OR E138A**
- RAL – High Resistance
- EVG – High Resistance
- DTG – Intermediate Resistance
- BIC – Intermediate Resistance

→ **Q148R + G140A + E138A**
- RAL – High Resistance
- EVG – High Resistance
- DTG – High Resistance
- BIC – High Resistance

N155H
- RAL – High Resistance
- EVG – High Resistance
- DTG – Susceptible
- BIC – Susceptible

→ **N155 + E92Q**
- RAL – High Resistance
- EVG – High Resistance
- DTG – Intermediate Resistance
- BIC – Intermediate Resistance

Patient Case #1

- 38yo male with HIV diagnosis in July 2007
- Started on Atripla (efavirenz/emtricitabine/TDF)
- Undetectable viral load from 2007 – 2015
- Started missing doses in 2015 due to change in work schedule

	11/2014	3/2015	7/2015	9/2015
CD4	494	382	364	308
VL	< 40	1,542	3,664	2,774

- Genotype ordered

Patient Case 1

- Genotype shows: M184V, K103N
  - High level resistance to emtricitabine (NRTI) and efavirenz (NNRTI)
- What regimen could we start?

Patient Case 2

- 59 year old male in care since 1995
- Significant ART history
  - DDI, 3TC, IDV – transferred to clinic on regimen
  - DDI, ABC, 3TC, EFV – 2000
  - TDF, ABC, LPV/r – 2002
  - TDF, LPV/r – 2009 (patient self d/c'd ABC)
  - ABC, TDF, LPV/r – 2009
- Long history of non-adherence leading to resistance and regimen changes

Patient Case 2

- Patient presents back to clinic after lost to follow-up for ~6 months
- Still sporadically taking a supply of last regimen – ABC, TDF, LPV/r
- Patient has a detectable viral load so a genotype is ordered

Patient Case 2

- Genotype shows:
  - RT: 74V, 75A/I/T, 100I, 103N, 115F, 151K/L/M, 184V
  - P: 10F, 15V, 20M, 36I, 54V, 63P, 71V, 72V, 82A/I/T, 85V
- Interpretation:
  - High level resistance to all NRTIs save TDF which is intermediate
  - High level resistance to all NNRTIs
  - High level resistance to all PIs save for DRV/r which is susceptible and ATV, FPV, SQV which are intermediate
  - No data on INSTIs or CCR5

Patient Case 2

- What options are left?
- What regimen should the patient receive?
- What factors are importance before starting patient back on a new regimen?

Patient Case 3

- 23 year old male newly diagnosed in April 2015
- Baseline genotype showed no mutations
- Started on a regimen of Complera (emtricitabine/tenofovir/rilpivirine)

Date	Baseline	6/15	9/15
CD4	402	358	339
VL	195,302	130,980	141,429

- What mutations might you expect from a genotype?

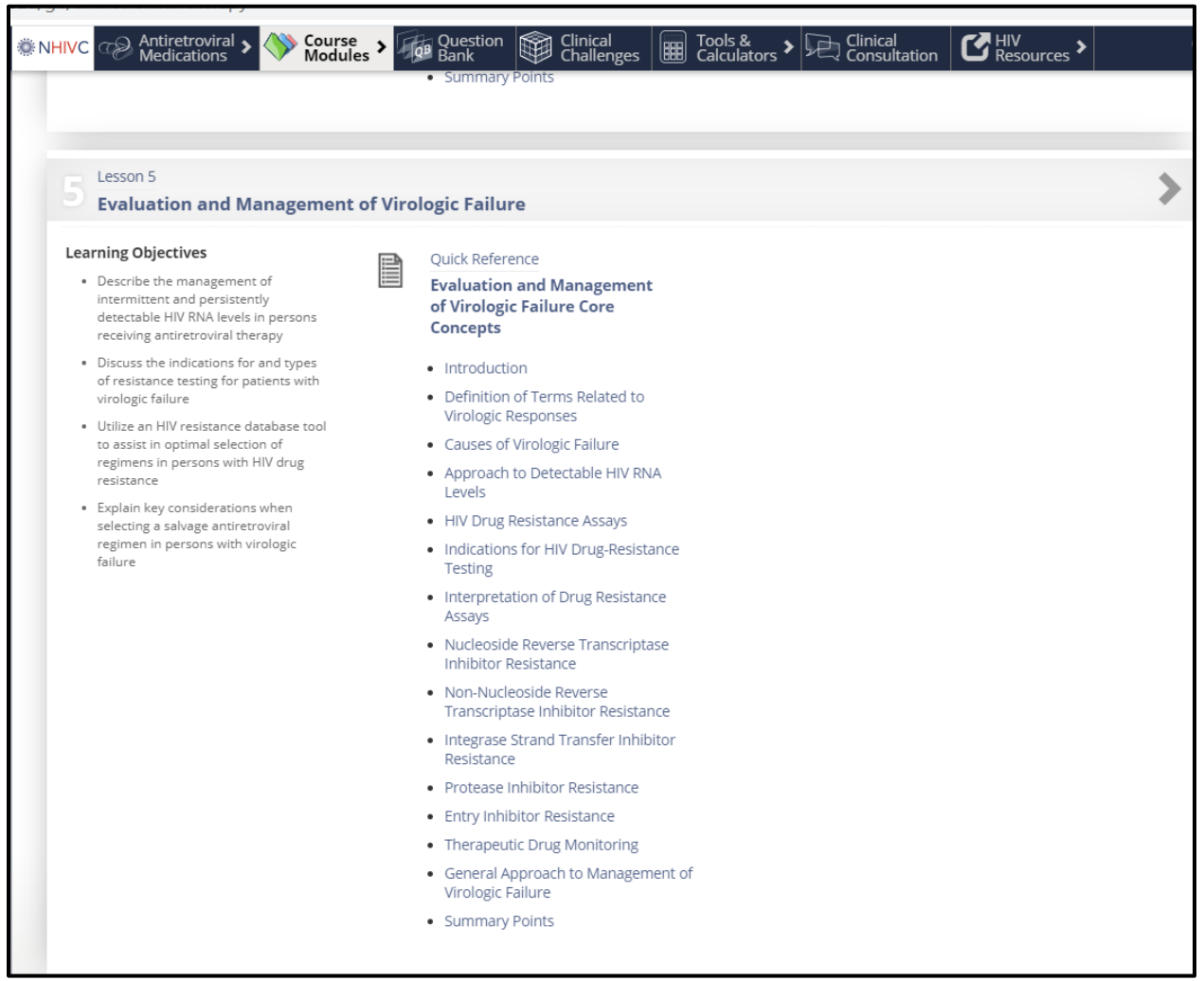
Patient Case 3

- Genotype shows:
  - PI: M36I, I62V, L63P, A71V, V77I, I93L
  - All mutations are polymorphic = virus is wild type
- Recommendation:
  - Address adherence

Resistance Resources

- Stanford HIV Database
  - http://hivdb.stanford.edu/
- IAS-USA
  - https://www.iasusa.org/resources/hiv-drug-resistance-mutations/
- Clavel F, Hance AJ. HIV Drug Resistance. N Engl J Med 2004;350:1023-35.
- DHHS, *Guidelines for the Use of Antiretroviral Agents in HIV-1-Infected Adults and Adolescents*. https://clinicalinfo.hiv.gov

For more information…



https://www.hiv.uw.edu/go/antiretroviral-therapy

Thank you!

Questions?

mforsyth@wayne.edu

Questions answered within 24 – 48 hours

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