

# Drug resistance epidemiology and clinical use of resistance tests in HIV

*Carlo Federico Perno*



*23 November 2013*

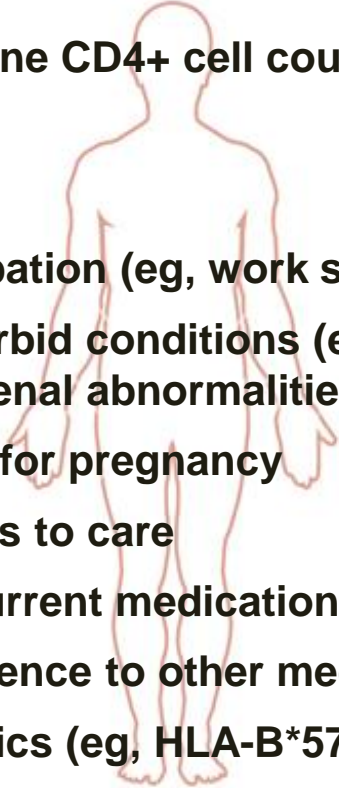
# 2013 Recommendations for ART Initiation

- ART is recommended for *all* HIV-infected ART-naive pts to reduce risk of disease progression and transmission
  - Strength of recommendation varies by CD4+ cell count and risk group (perinatal, heterosexual, other)
  - Pts should be ready to commit to ART and understand benefits and risks of therapy and importance of adherence; individual pts may elect to defer ART
- **Selection of a regimen should be individualized** on the basis of virologic efficacy, toxicity, pill burden, dosing frequency, drug–drug interaction potential, resistance testing results, and comorbid conditions

# Considerations When Selecting First-line Antiretroviral Therapy

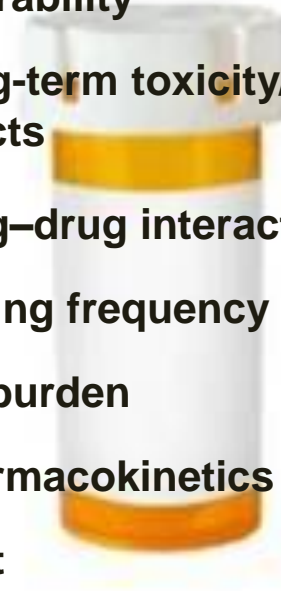
## Patient/Viral Factors

- Baseline HIV-1 RNA level
- Baseline CD4+ cell count
- Age
- Sex
- Occupation (eg, work schedule)
- Comorbid conditions (eg, CV risk, renal abnormalities)
- Plans for pregnancy
- Access to care
- Concurrent medications
- Adherence to other medications
- Genetics (eg, HLA-B\*5701)
- Viral tropism



## Antiretroviral Drug Factors

- Efficacy
- Baseline drug resistance
- Tolerability
- Long-term toxicity/metabolic effects
- Drug–drug interactions
- Dosing frequency
- Pill burden
- Pharmacokinetics
- Cost



**All the current guidelines agree that  
the primary goal of antiretroviral therapy  
is to suppress HIV RNA maximally  
(**<20–75 copies/mL**, depending on the assay used)  
in order to preserve immunologic function and  
increase disease-free survival**

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

***February, 2013***



Developed by the HHS Panel on Antiretroviral Guidelines for Adults  
and Adolescents – A Working Group of the Office of AIDS Research  
Advisory Council (OARAC)

**Antiretroviral Treatment of Adult HIV Infection**

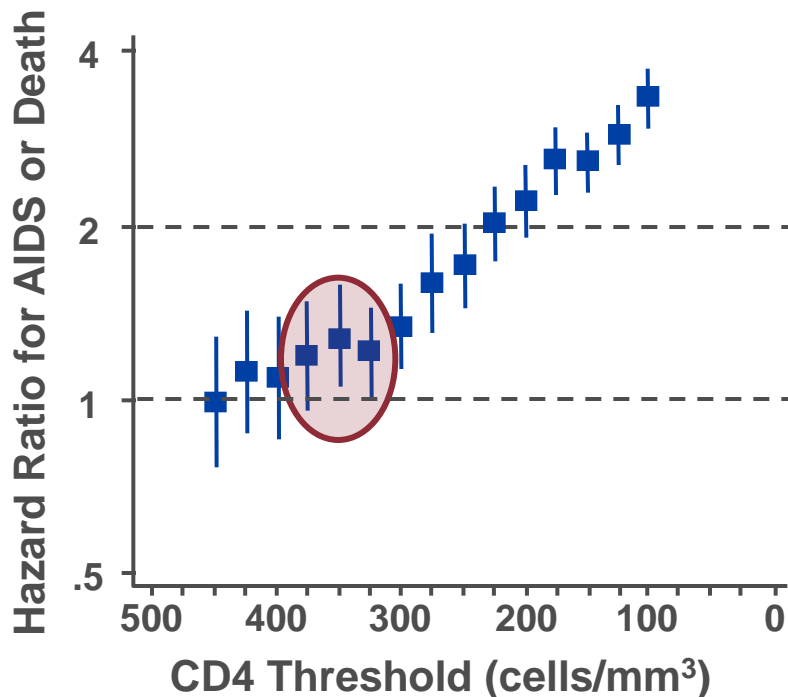
2012 Recommendations of the International  
Antiviral Society–USA Panel

***July 2012***

**Guidelines for the Clinical  
Management and Treatment  
of HIV Infected Adults in Europe  
European AIDS Clinical Society  
(EACS) *October 2013***

# ART-CC: Delay in starting ART is associated with an increased risk of AIDS or death

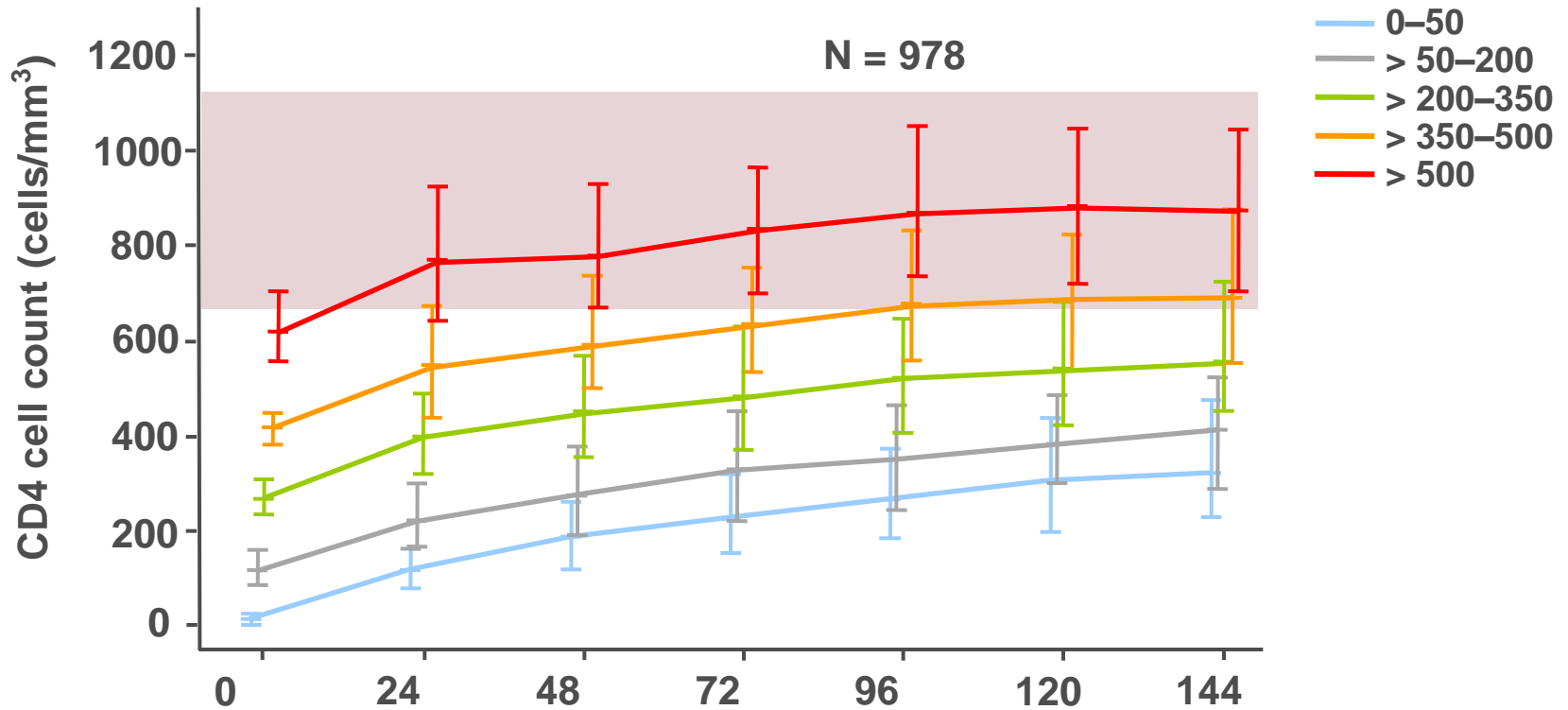
Hazard ratios for AIDS or death, adjusted for lead time/unseen events



Comparison	Hazard Ratio (95% CI)
276–375 vs 376–475	1.19 (0.96 to 1.47)
251–350 vs 351–450	1.28 (1.04 to 1.57)
226–325 vs 326–425	1.21 (1.01 to 1.46)

**Delaying ART to <350 (but not <375) cells/mm<sup>3</sup> is associated with an increased risk of AIDS or death**

# ACTG 384: Immunological recovery over time



After 3 years' follow-up, patients with baseline CD4 cell counts >350 cells/mm<sup>3</sup> approached those of uninfected reference individuals (pink box); patients with baseline CD4 <350 cells/mm<sup>3</sup> generally did not regain normal counts

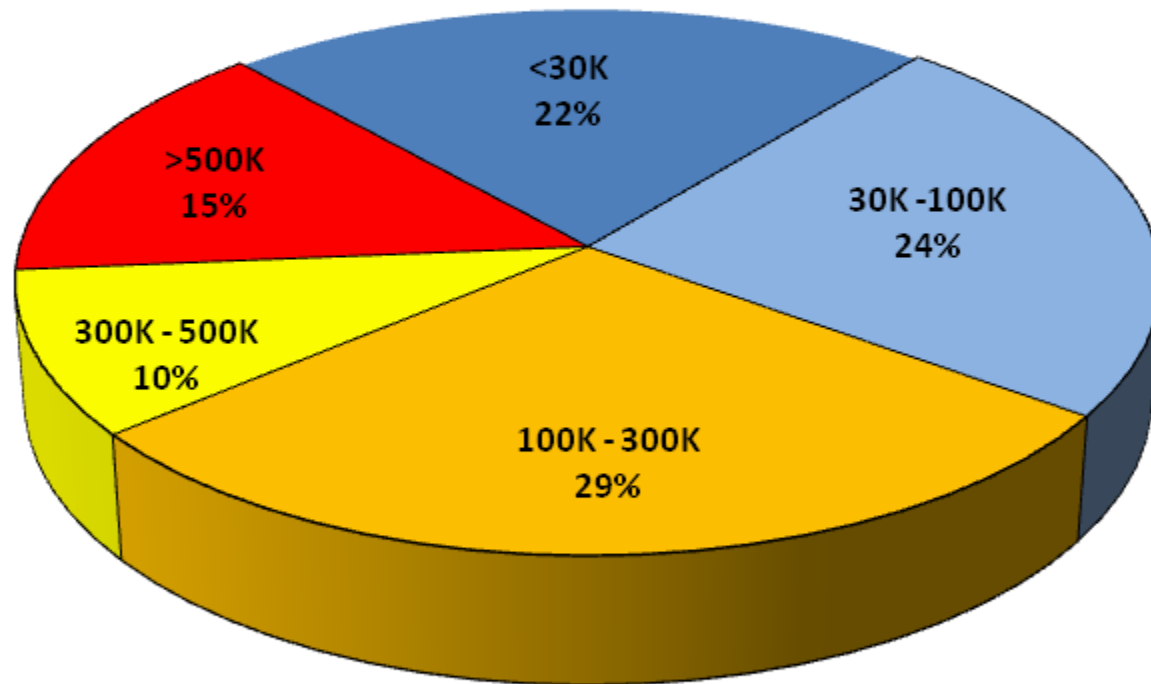
**✓ During combination antiretroviral therapy, virological undetectability is frequently achieved.**

**..even if at different times!**

**..Indeed....**

# About a quarter of patients has pre-HAART viremia >300K copies/ml

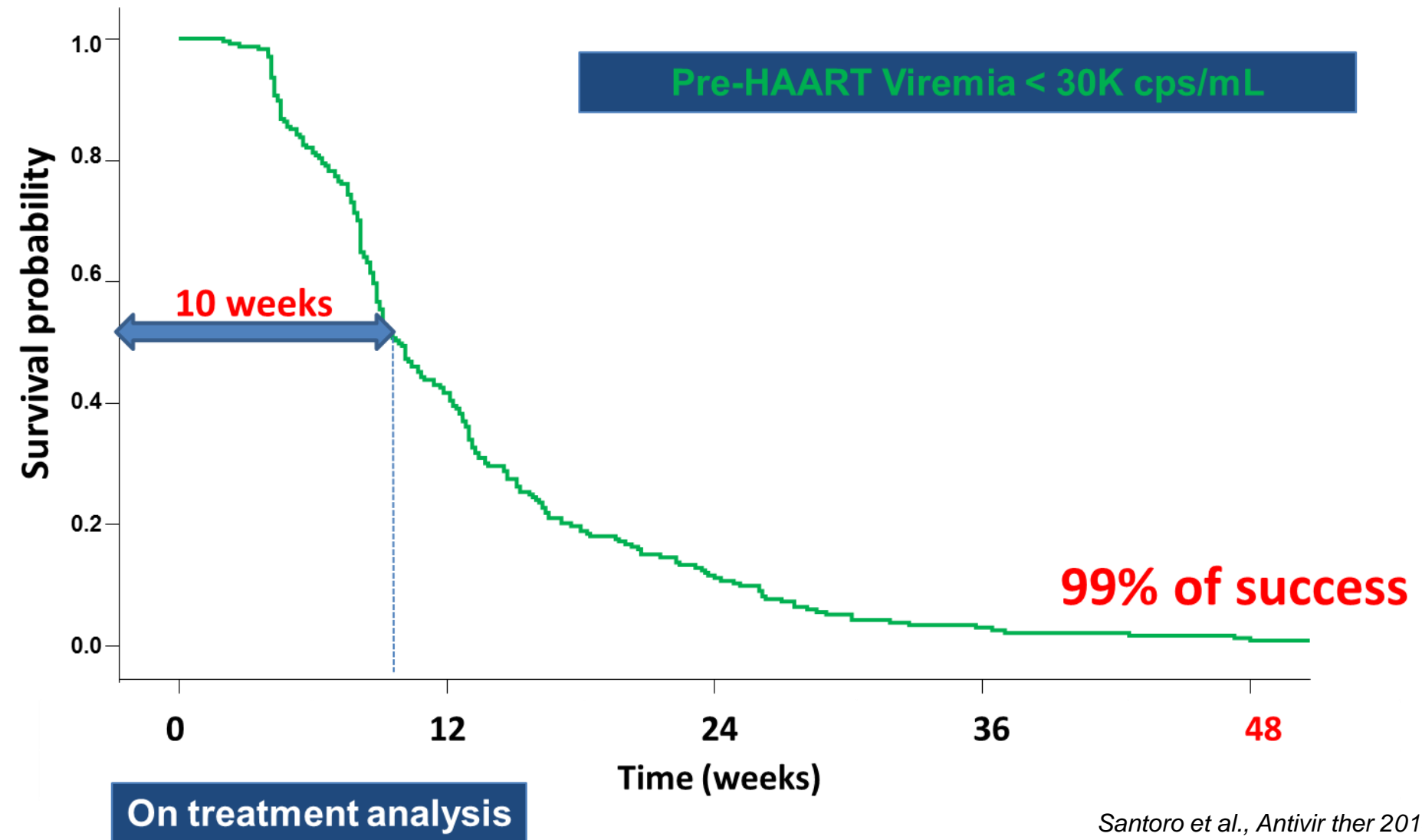
Pre-HAART viremia (copies/mL) stratifications





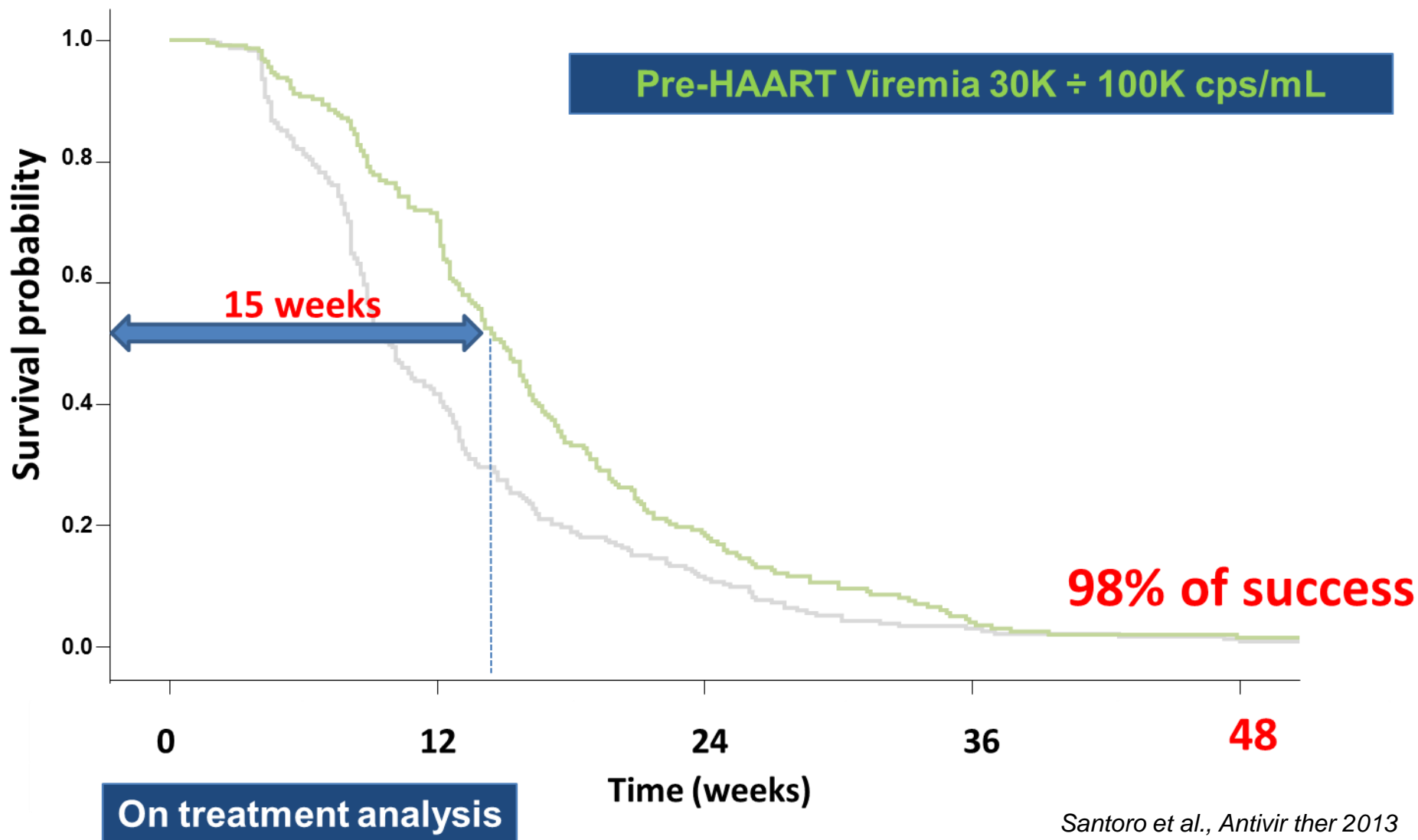
# The time to achieve virological undetectability and the rate of success at 48 week are pre-HAART viremia dependent

Kaplan Meyer survival curves (Event viremia < 50 copies/mL)



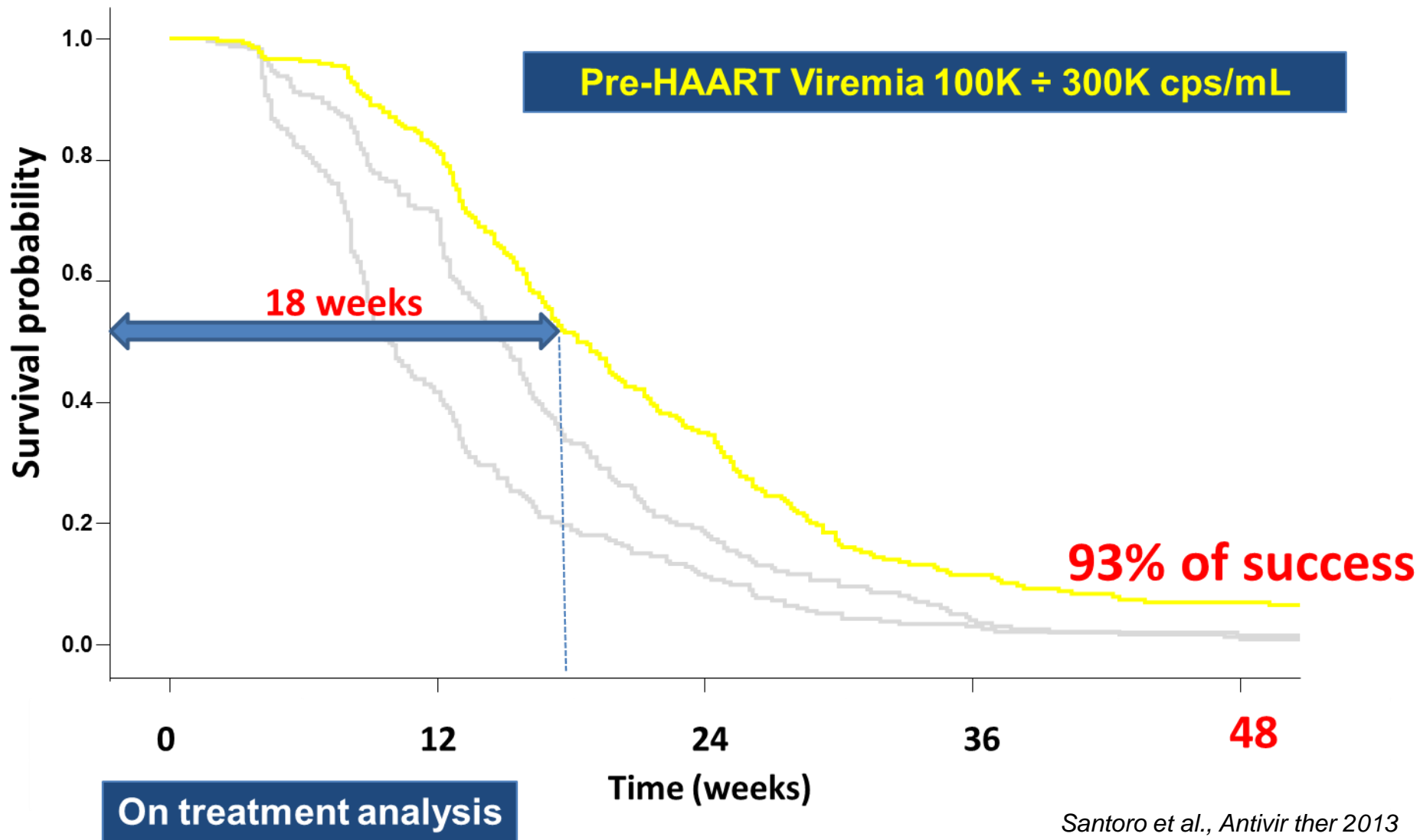
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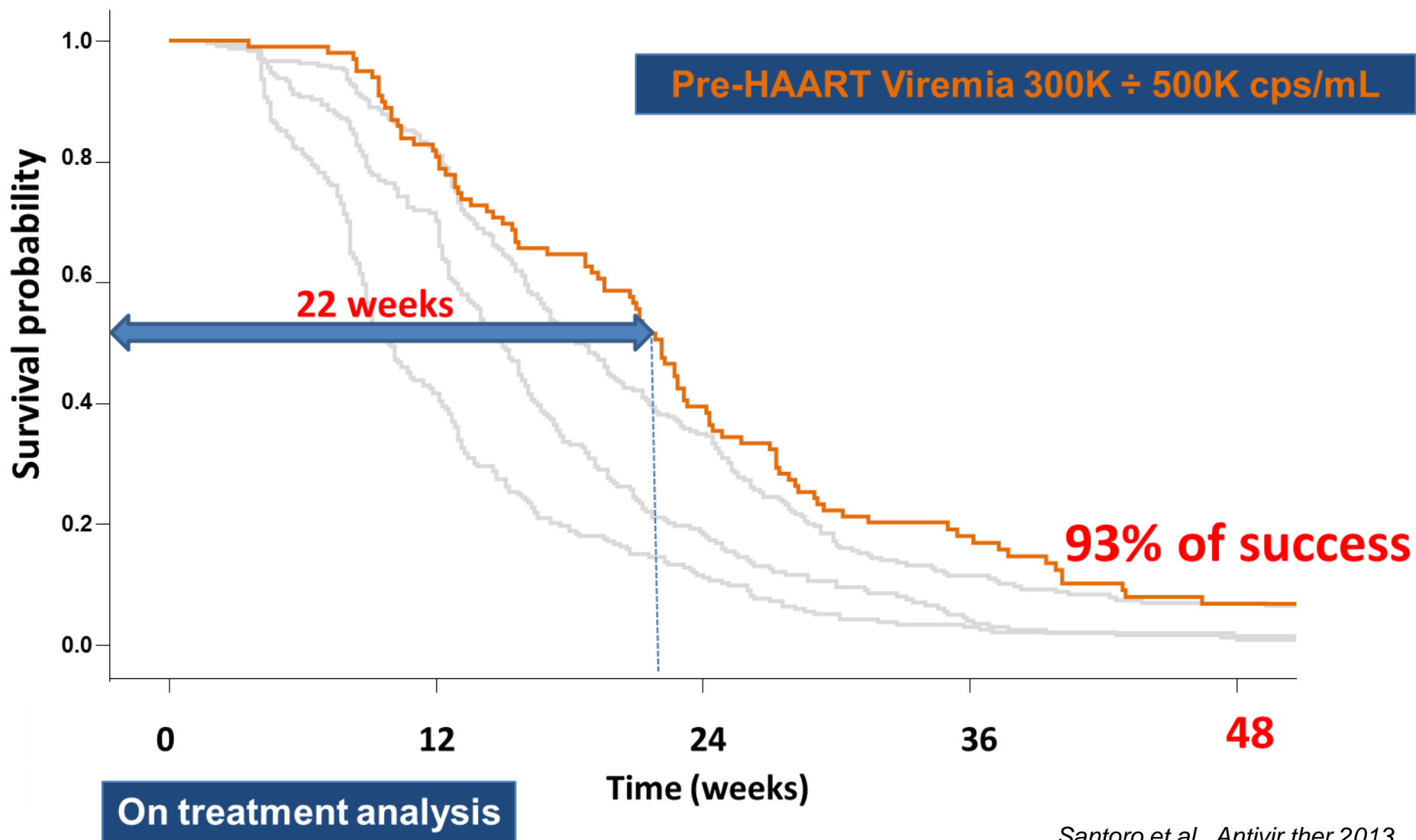
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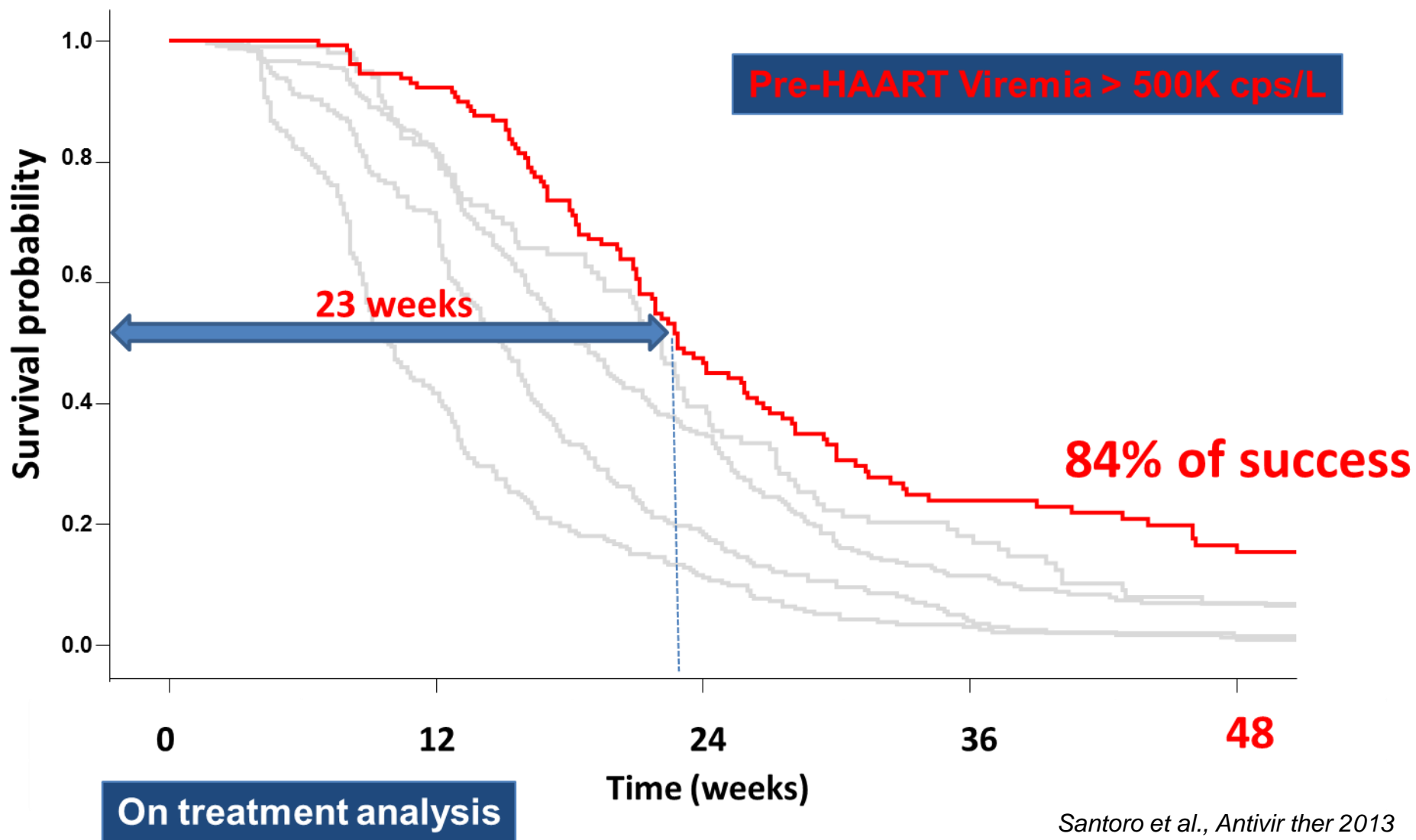
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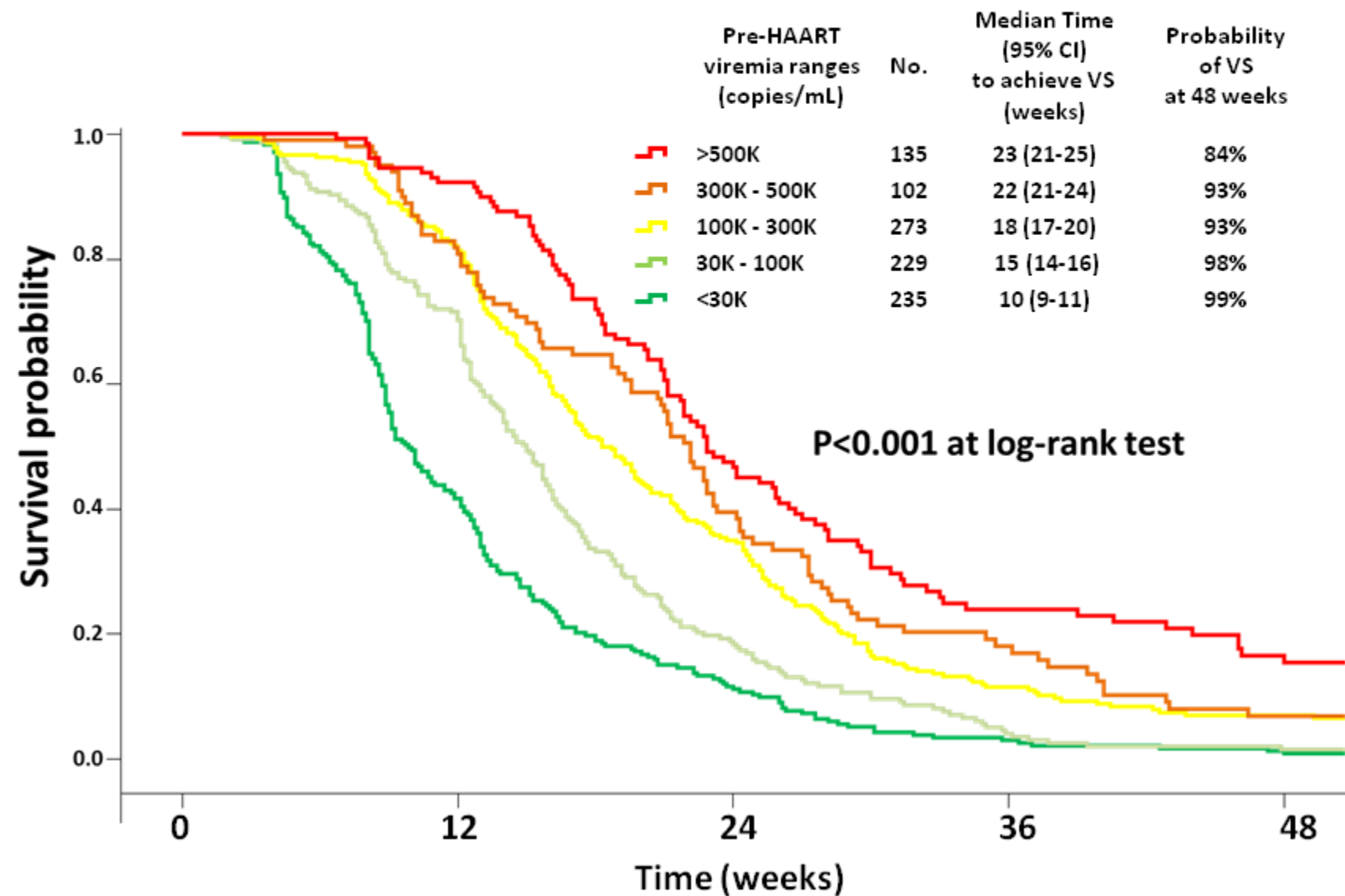


# The time to achieve virological undetectability and the rate of success at 48 week are pre-HAART viremia dependent

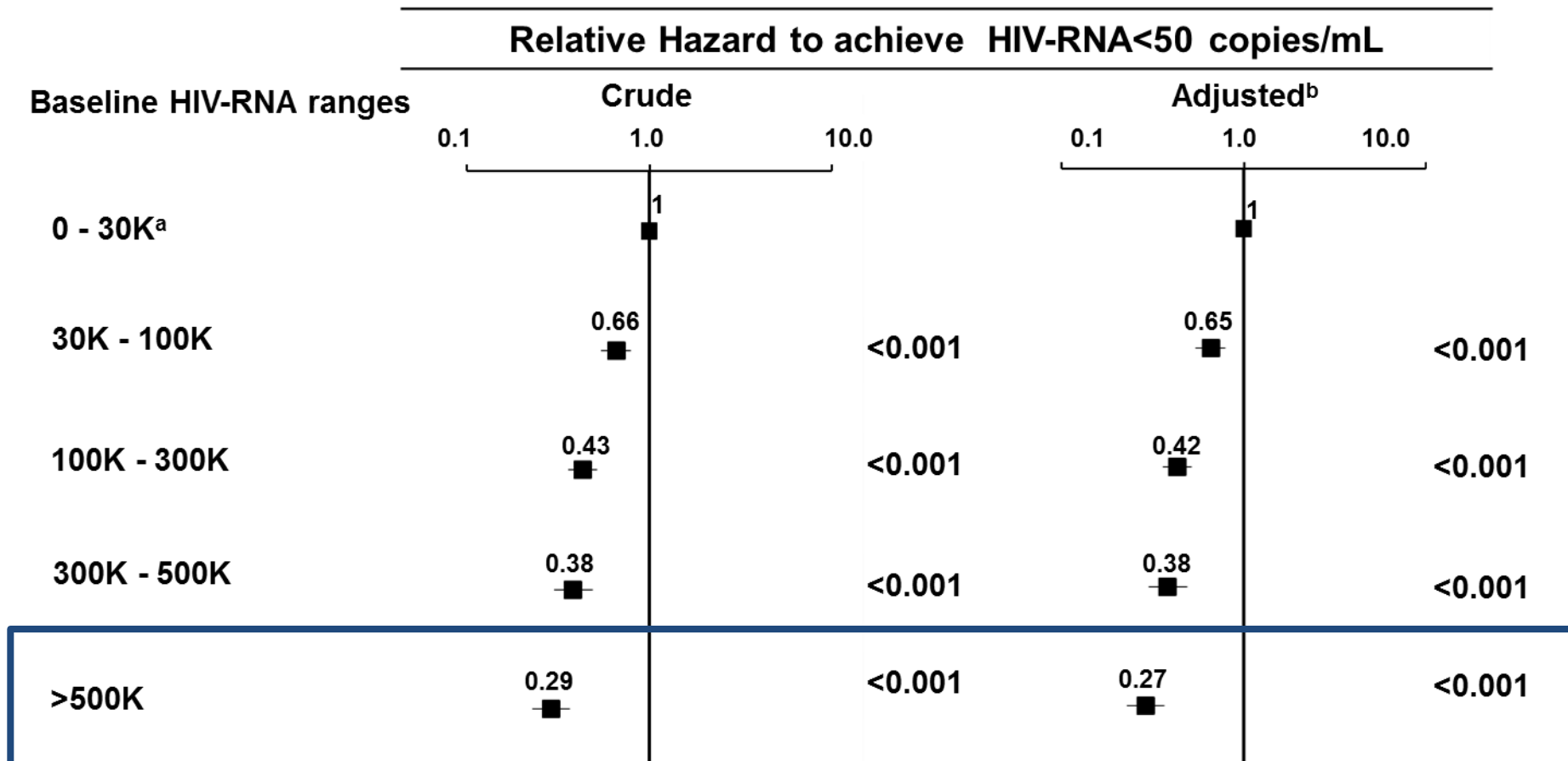
Kaplan Meyer survival curves (Event viremia < 50 copies/mL)



# The time to achieve virological undetectability and the rate of success at 48 weeks are pre-HAART viremia dependent



Patients having **pre-HAART viremia >500K copies/mL** have the lowest relative hazard to achieve virological success in comparison to other ones, also after adjusting for gender, age, pre-HAART CD4 cell count, transmitted drug resistance, calendar year and third drug administered (PI/r vs. NNRTI)



<sup>a</sup> Reference range.

<sup>b</sup> Adjusted for: gender, age, pre-HAART CD4 cell count, transmitted drug resistance, calendar year, third drug (PI vs. NNRTI).

# Virologic Failure Following Persistent Low-level Viremia in a Cohort of HIV-Positive Patients: Results From 12 Years of Observation

Claudie Laprise,<sup>1</sup> Alexandra de Pokomandy,<sup>2,3</sup> Jean-Guy Baril,<sup>4</sup> Serge Dufresne,<sup>4</sup> and Helen Trottier<sup>1</sup>

<sup>1</sup>Department of Social and Preventive Medicine, University of Montreal, Sainte-Justine Hospital Research Center, <sup>2</sup>Department of Family Medicine, McGill University, <sup>3</sup>Chronic Viral Illnesses Service, McGill University Health Center, and <sup>4</sup>Clinique Médicale du Quartier Latin, Montreal, Canada

**Background.** The current goal of antiretroviral therapy (ART) is to maintain a suppressed human immunodeficiency virus (HIV) viral load below limits of assay detection. When viral loads remain in low-level viremia (LLV), especially between 50 and 200 copies/mL, the best management and clinical consequences remain unknown. Our objective was to study the long-term **impact of persistent LLV on the subsequent risk of virologic failure** in a cohort of people living with HIV in Montreal, Canada.

**Methods.** We compared the cumulative **incidence of subsequent virologic failure** (defined as an HIV RNA viral load of >1000 copies/mL) in patients receiving ART for at least 12 months by following **4 persistence categories (<50, 50–199, 200–499, and 500–999 copies/mL)** for 6, 9, or 12 months, using Kaplan-Meier analysis. The association between subsequent virologic failure and persistence status were estimated using a Cox proportional hazards model.

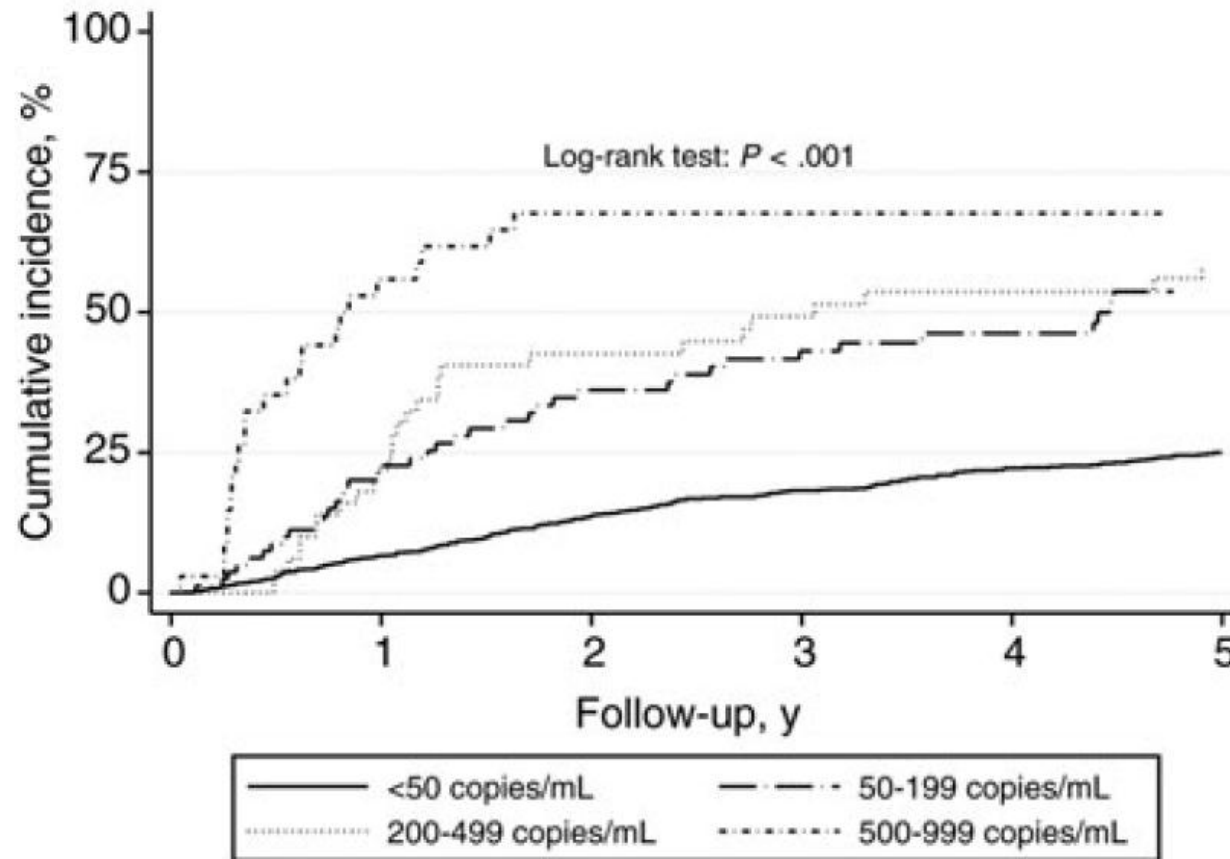
**Results.** The cumulative incidence of virologic failure 1 year after having maintained a LLV for 6 months was 22.7% (95% confidence interval [CI], 14.9–33.6) for 50–199 copies/mL, 24.2% (95% CI, 14.5–38.6) for 200–499 copies/mL, and 58.9% (95% CI, 43.1–75.2) for 500–999 copies/mL, compared with 6.6% (95% CI, 5.3–8.2) for an undetectable HIV RNA viral load. Even after adjustment for potential confounders, a persistent LLV of 50–199 copies/mL for 6 months doubled the risk of virologic failure (hazard ratio, 2.22; 95% CI, 1.60–3.09), compared with undetectable viral loads for the same duration. Similar results have been found for persistent LLV of 9 or 12 months.

**Conclusions.** **In this cohort, all categories of persistent LLV between 50 and 999 copies/mL were associated with an increased risk of virologic failure. The results shed new light for the management of patients with LLV, especially with regard to LLV of 50–199 copies/mL.**



The cumulative incidences of subsequent virologic failure (ie, > 1000 copies/mL) over 5 years, following persistence of LLV was significantly higher for all LLV strata compared to who maintained undetectable HIV load.

Persistent viral load status defined with a duration of 6 months



# Viremia Copy-Years Predicts Mortality Among Treatment-Naive HIV-Infected Patients Initiating Antiretroviral Therapy

*AIDS* 2011;53:927–935

**Michael J. Mugavero,<sup>1,a</sup> Sonia Napravnik,<sup>2,3,a</sup> Stephen R. Cole,<sup>3</sup> Joseph J. Eron,<sup>2,3</sup> Bryan Lau,<sup>4</sup> Heidi M. Crane,<sup>5</sup> Mari M. Kitahata,<sup>5</sup> James H. Willig,<sup>1</sup> Richard D. Moore,<sup>6</sup> Steven G. Deeks,<sup>7</sup> Michael S. Saag,<sup>1</sup> and on behalf of the Centers for AIDS Research Network of Integrated Clinical Systems (CNICS) Cohort Study**

**Background.** Cross-sectional plasma human immunodeficiency virus (HIV) viral load (VL) measures have proven invaluable for clinical and research purposes. However, cross-sectional VL measures fail to capture cumulative plasma HIV burden longitudinally. We evaluated the cumulative effect of exposure to HIV replication on mortality following initiation of combination antiretroviral therapy (ART).

**Methods.** We included treatment-naive HIV-infected patients starting ART from 2000 to 2008 at 8 Center for AIDS Research Network of Integrated Clinical Systems sites. Viremia copy-years, a time-varying measure of cumulative plasma HIV exposure, were determined for each patient using the area under the VL curve. Multivariable Cox models were used to evaluate the independent association of viremia copy-years for all-cause mortality.

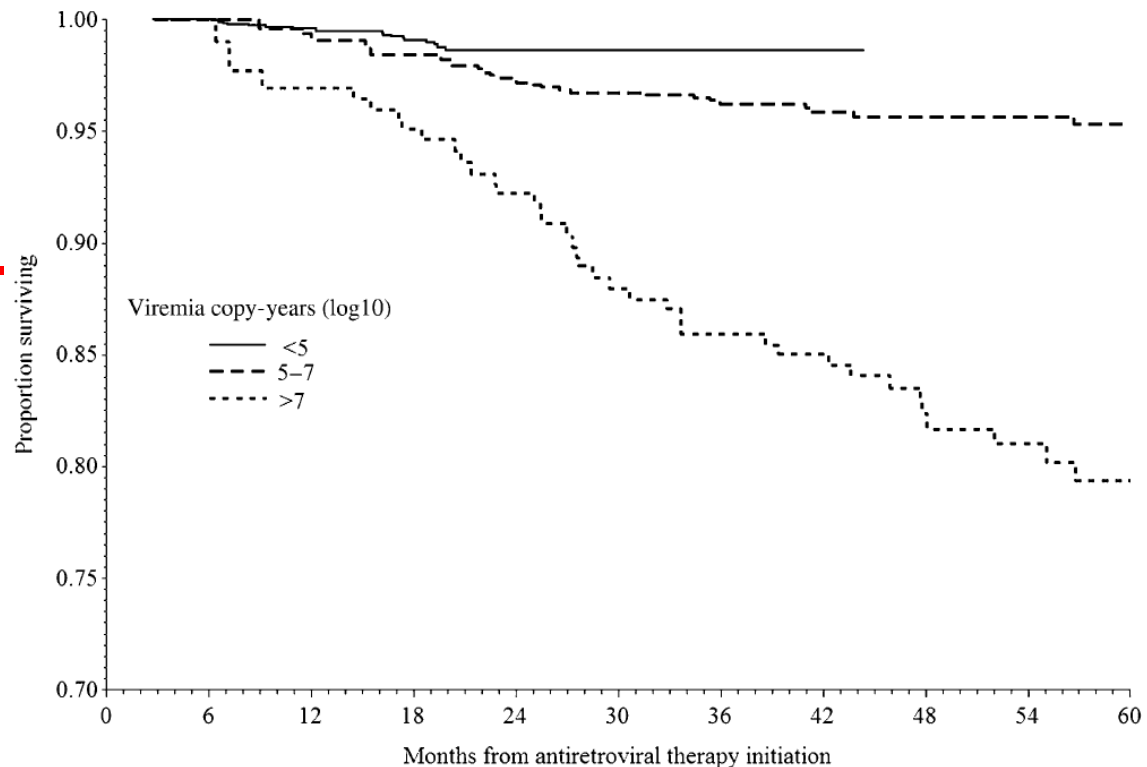
**Results.** Among 2027 patients contributing 6579 person-years of follow-up, the median viremia copy-years was  $5.3 \log_{10} \text{ copy} \times \text{y/mL}$  (interquartile range:  $4.9\text{--}6.3 \log_{10} \text{ copy} \times \text{y/mL}$ ), and 85 patients (4.2%) died. When evaluated separately, viremia copy-years (hazard ratio [HR] = 1.81 per  $\log_{10} \text{ copy} \times \text{y/mL}$ ; 95% confidence interval [CI], 1.51–2.18 per  $\log_{10} \text{ copy} \times \text{y/mL}$ ), 24-week VL (1.74 per  $\log_{10} \text{ copies/mL}$ ; 95% CI, 1.48–2.04 per  $\log_{10} \text{ copies/mL}$ ), and most recent VL (HR = 1.89 per  $\log_{10} \text{ copies/mL}$ ; 95% CI: 1.63–2.20 per  $\log_{10} \text{ copies/mL}$ ) were associated with increased mortality. When simultaneously evaluating VL measures and controlling for other covariates, viremia copy-years increased mortality risk (HR = 1.44 per  $\log_{10} \text{ copy} \times \text{y/mL}$ ; 95% CI, 1.07–1.94 per  $\log_{10} \text{ copy} \times \text{y/mL}$ ), whereas no cross-sectional VL measure was independently associated with mortality.

**Conclusions.** Viremia copy-years predicted all-cause mortality independent of traditional, cross-sectional VL measures and time-updated CD4+ T-lymphocyte count in ART-treated patients, suggesting cumulative HIV replication causes harm independent of its effect on the degree of immunodeficiency.

# Viremia Copy-Years Predicts Mortality Among Treatment-Naive HIV-Infected Patients Initiating Antiretroviral Therapy

**Cumulative plasma HIV burden, demonstrated prognostic value for all-cause mortality among 2027 HIV-infected patients following ART**

**(viral load values prior to 24 weeks of ART initiation were excluded)**



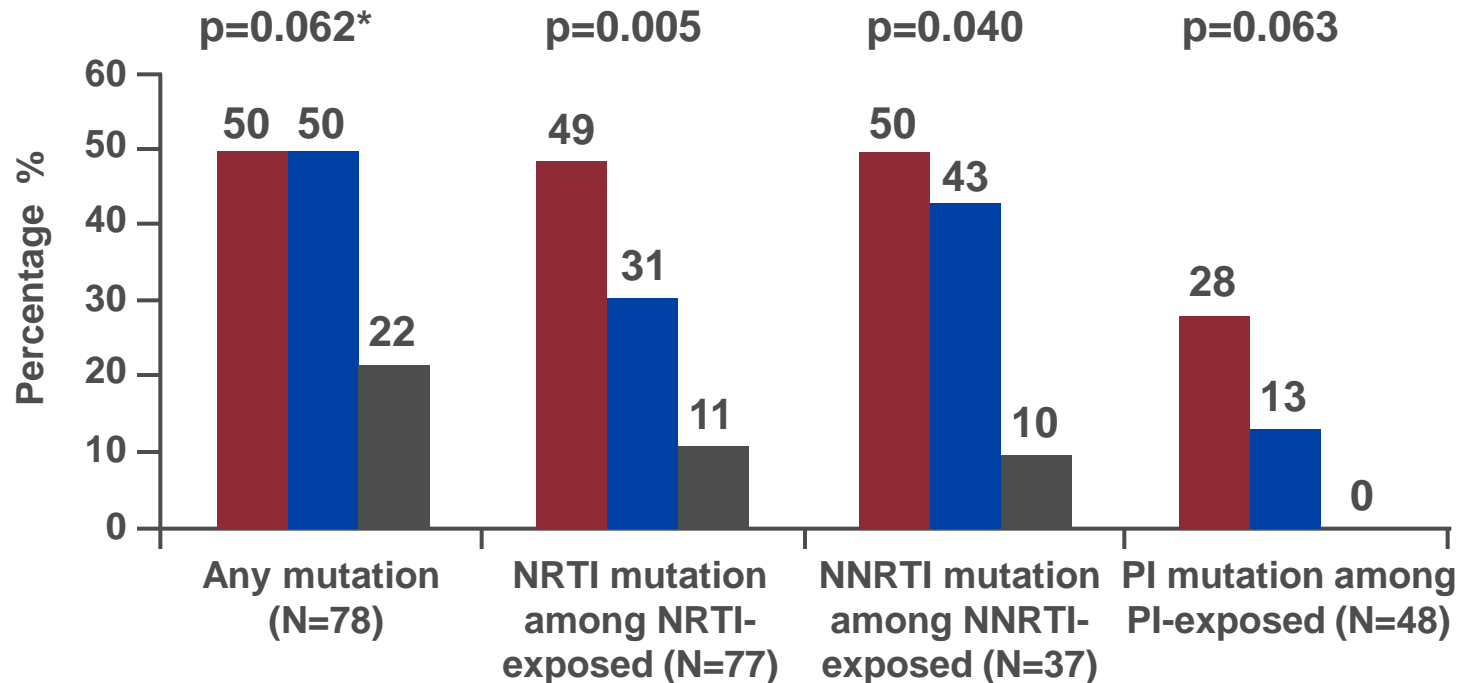
Viremia copy-years (log <sub>10</sub> )	Number at risk									
	6 mo	12 mo	18 mo	24 mo	30 mo	36 mo	42 mo	48 mo	54 mo	60 mo
<5	1692	1414	1105	317	79	18	2			
5-7	245	289	332	833	851	727	606	509	414	316
>7	90	135	181	192	183	169	152	132	122	108

- **Obtaining and maintaining undetectable viral level is crucial for long-term success**

- **Avoiding (and properly managing) drug resistance helps in achieving this clinical result**

# Initiating HAART earlier reduces the development of resistance at virological failure

Frequency of genotype mutations among persons with virologic failure after successful suppression on HAART, by CD4 cell count at HAART (N=78), the HOPS, 1999–2006



CD4 stratum: ■ 0–199 cells/mm<sup>3</sup> ■ 200–349 cells/mm<sup>3</sup> ■ >350 cells/mm<sup>3</sup>

HOPS = HIV Outpatient Study

- *Virus continues to evolve if kept under pressure of failing antiviral therapy...*

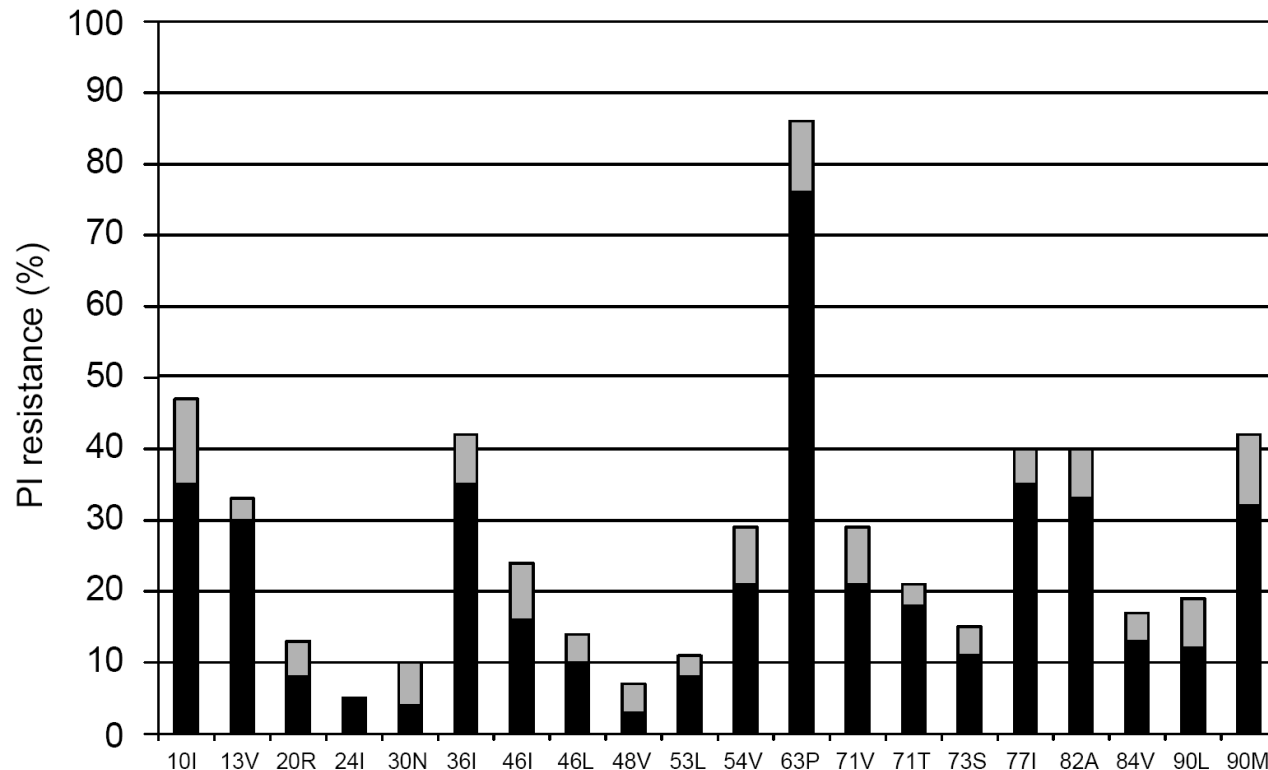
This may increase cross-resistance, and then decrease chances of efficacy of subsequent drugs and regimens.

In the frame of a correct therapeutic sequencing, first failing therapies should be changed as soon as possible after definition of virological failure.

# Evolution of drug resistance in patients remaining on a virologically failing cART

110 patients on failing cART for a median of 11 months (range 6-50)

- Overall, 6-monthly increase of 1.96 IAS mutations, and an average loss of 1.25 active drugs were estimated





# Impact of Low-Level-Viremia on HIV-1 Drug-Resistance Evolution among Antiretroviral Treated-Patients

Constance Delaugerre<sup>1,2,3\*</sup>, Sébastien Gallien<sup>2,3,4</sup>, Philippe Flandre<sup>5,6</sup>, Dominique Mathez<sup>7</sup>, Rishma Amarsy<sup>1</sup>, Samuel Ferret<sup>4</sup>, Julie Timsit<sup>8</sup>, Jean-Michel Molina<sup>2,3,4</sup>, Pierre de Truchis<sup>9</sup>



## Abstract

**Background:** Drug-resistance mutations (DRAM) are frequently selected in patients with virological failure defined as viral load (pVL) above 500 copies/ml (c/mL), but few resistance data are available at low-level viremia (LLV). **Our objective** was to determine the emergence and evolution of DRAM during LLV in HIV-1-infected patients while receiving antiretroviral therapy (ART).

**Methods:** Retrospective analysis of patients presenting a LLV episode defined as pVL between 40 and 500 c/mL on at least 3 occasions during a 6-month period or longer while on the same ART. Resistance genotypic testing was performed at the onset and at the end of LLV period. Emerging DRAM was defined during LLV if never detected on baseline genotype or before.

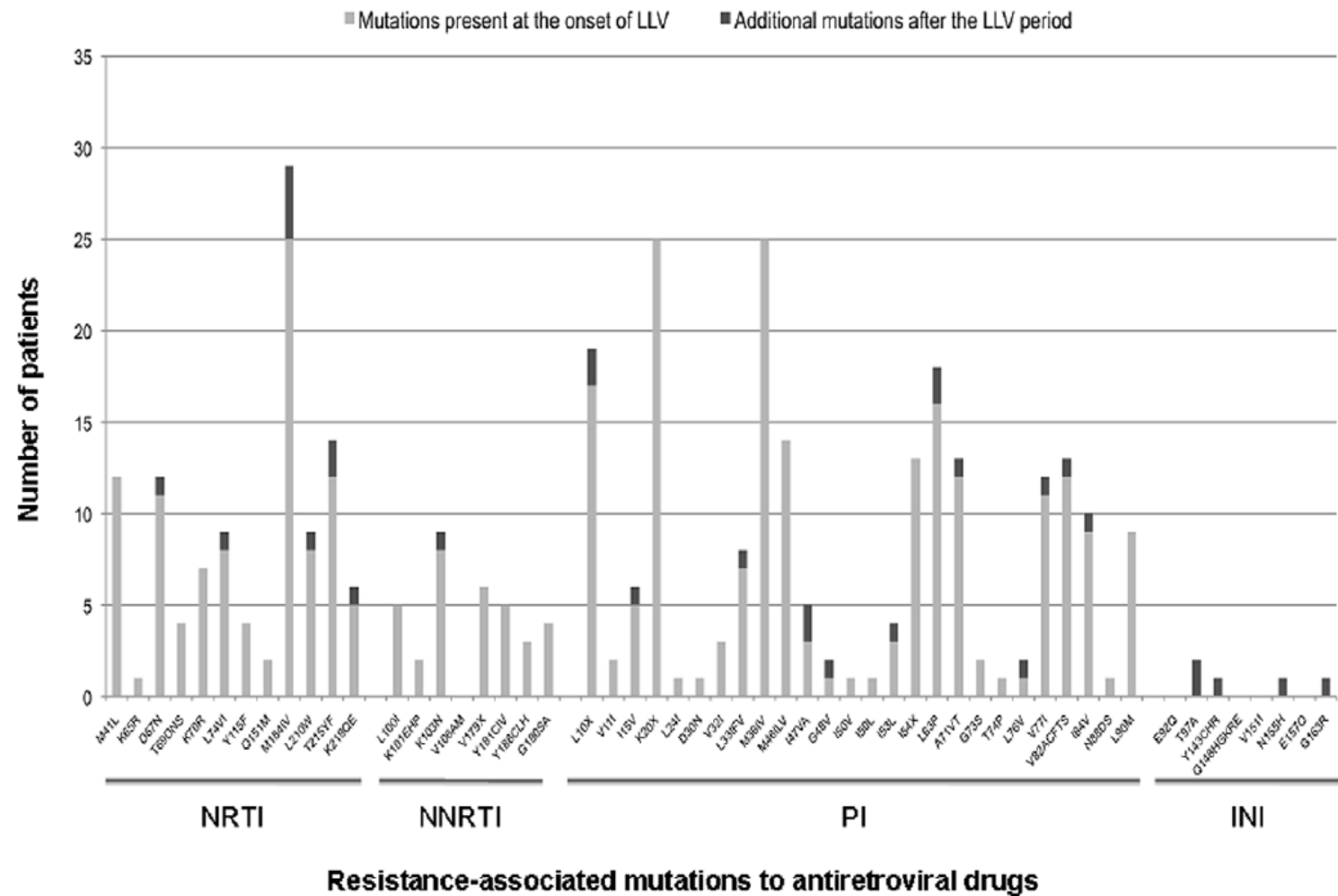
**Results:** 48 patients including 4 naive and 44 pretreated (median 9 years) presented a LLV episode with a median duration of 11 months. Current ART included 2NRTI (94%), ritonavir-boosted PI (94%), NNRTI (23%), and/or raltegravir (19%). Median pVL during LLV was 134 c/mL. Successful resistance testing at both onset and end of the LLV episode were obtained for 37 patients (77%), among who 11 (30%) acquired at least 1 DRAM during the LLV period: for NRTI in 6, for NNRTI in 1, for PI in 4, and for raltegravir in 2. During the LLV period, number of drugs with genotypic resistance increased from a median of 4.5 to 6 drugs. Duration and pVL level of LLV episode, duration of previous ART, current and nadir CD4 count, number of baseline DRAM and GSS were not identified as predictive factors of resistance acquisition during LLV, probably due to limited number of patients.

**Conclusion:** Persistent LLV episodes below 500 c/ml while receiving ART is associated with emerging DRAM for all drug classes and a decreasing in further therapeutic options, suggesting to earlier consider resistance monitoring and ART optimization in this setting.



# Impact of Low-Level-Viremia on HIV-1 Drug-Resistance Evolution among Antiretroviral Treated-Patients

Constance Delaugerre<sup>1,2,3\*</sup>, Sébastien Gallien<sup>2,3,4</sup>, Philippe Flandre<sup>5,6</sup>, Dominique Mathez<sup>7</sup>, Rishma Amarsy<sup>1</sup>, Samuel Ferret<sup>4</sup>, Julie Timsit<sup>8</sup>, Jean-Michel Molina<sup>2,3,4</sup>, Pierre de Truchis<sup>9</sup>



# Multiple drug class-wide resistance associated with poorer survival after treatment failure in a cohort of HIV-infected patients

Mauro Zaccarelli<sup>a</sup>, Valerio Tozzi<sup>a</sup>, Patrizia Lorenzini<sup>a</sup>, Maria P. Trotta<sup>a</sup>, Federica Forbici<sup>b</sup>, Ubaldo Visco-Comandini<sup>a</sup>, Caterina Gori<sup>b</sup>, Pasquale Narciso<sup>a</sup>, Carlo F. Perno<sup>b</sup> and Andrea Antinori<sup>a</sup> for the Collaborative Group for Clinical Use of HIV Genotype Resistance Test (GRT) at National Institute for Infectious Diseases 'Lazzaro Spallanzani'

**Objective:** To evaluate the effect of drug class-wide resistance (CWR) on survival in HIV-infected individuals who underwent genotypic resistance test after antiretroviral failure.

**Design:** Observational, longitudinal cohort study.

**Methods:** HIV-infected individuals experiencing treatment failure were enrolled at first genotypic resistance test. End-points were death for any cause, AIDS-related death and AIDS-defining event/death. CWR was defined according to the International AIDS Society consensus. Survival analysis was performed with Cox's model.

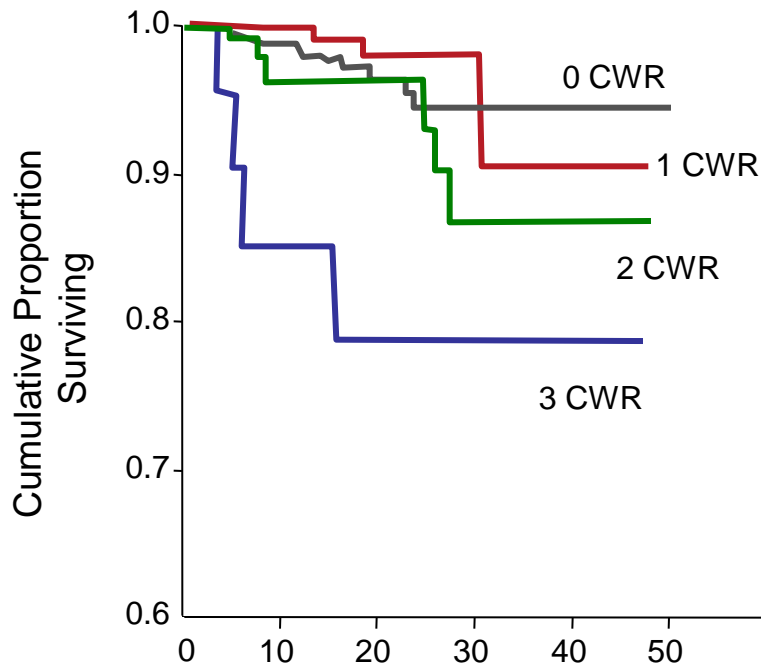
**Results:** Among 623 patients enrolled and followed for a median of 19 months (interquartile range, 12–29), Kaplan–Meier analyses for end-points at 48 months in patients with no CWR, one CWR, two CWR or three CWR were 8.9, 11.7, 13.4 and 27.1%, respectively, for death; 6.1, 9.9, 13.4 and 21.5%, respectively, for AIDS-related death; and 16.0, 17.7, 19.3 and 35.9%, respectively, for new AIDS event/death. In a multivariate Cox's model, higher HIV RNA level, previous AIDS and detection of three CWR (hazard ratio, 5.34; 95% confidence interval, 1.76–16.24) were all significantly associated with increased risk of death, while higher CD4 cell count and use of a new boosted protease inhibitor drug after identifying genotypic resistance were associated with reduced risk. Detection of three CWR was also significantly associated with higher risk of AIDS-related death and new AIDS event/death.

**Conclusions:** Even in the late era of highly effective antiretroviral treatments, detection of CWR, particularly if extended to all three drug classes is related to poorer clinical outcome and represents a risk-marker of disease progression and death.

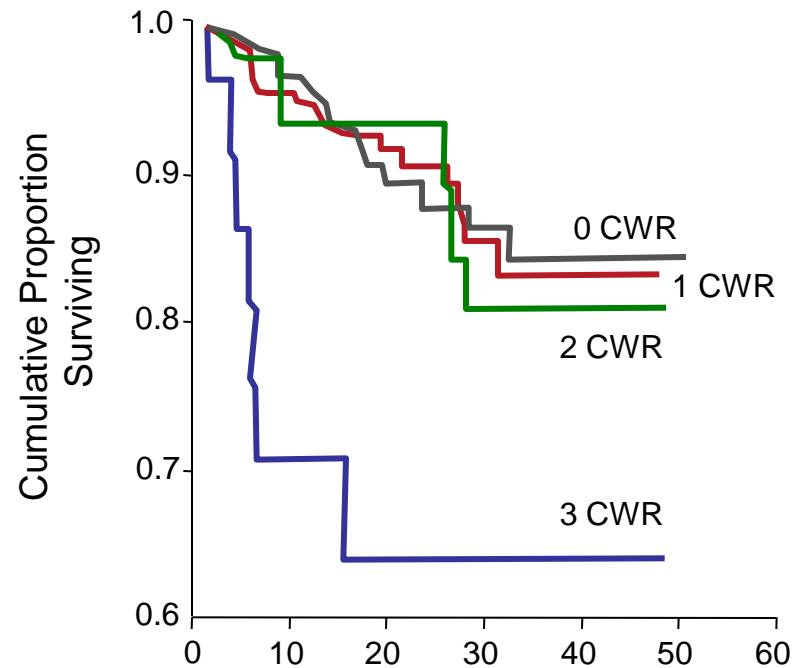
© 2005 Lippincott Williams & Wilkins

# Class-wide-resistance, Particularly if Extended to All 3 ARV Classes, Is Related to Poor Outcomes

Probability of surviving an AIDS related death



Probability of surviving/remaining free from AIDS events



**Resistance is irreversible and limits treatment options**

- Combination antiretroviral therapy to suppress HIV replication to below the levels of detection of sensitive plasma HIV RNA assays limits the potential for selection of antiretroviral resistant HIV variants and delays disease progression.

- **Therefore, maintaining an adequate pressure over the virus is crucial at anytime during antiviral therapy.**

Clinical Case: ID 7583 - Patient infected with HIV-1 B subtype

Age: 35

Sex: F

Risk Factor: unknown

CDC stage: C

in out

in out

- First seroconversion
- Drugs previously used

GRT July 2008  
PR: L63ST  
RT: L74LI V108VI  
T215TADN  
IN: none

GRT December 2010  
PR: : M46L L63P I84V I93L  
RT: A62V K103N M184V Y188L T215Y  
IN: N155H  
GP-120(V3): G15A E25D  
Tropism\*: Prevalence of a R5 tropic virus

**Virological failure after lowering genetic barrier by simplification to Raltegravir and Nevirapine**

IN: V31VIM I84VI T112AV  
M154I K211R

RT: V21I, D123E, T133L, D177E, G196E,  
T200I, A272P, E297K  
IN: S17N V31I I60M I84V M154I I208M K211R  
D288N

CD4 cell count (cells/mm<sup>3</sup>)

Viremia (log copies/ml)

From January 2005 to December 2006  
Therapy interruption

From January 2007 to July 2008:

TDF, FTC, FPV/r

On July 2008: VL 43361 cps/ml; CD4 350 cells/mm<sup>3</sup>

Undetectability threshold

Jul-08

Sep-08

Feb-09

Jun-09

Dec-09

Feb-10

May-10

Jul-10

Nov-10

Dec-10

Aug '06 - Jul '08  
FTC TDF FPV/r

Aug '08 (for 15 days)  
ddI FTC FPV/r

Aug '08 - Jul '10  
FPV/r RAL

Aug - Dec '10  
NVP RAL

# Resistance to antiviral drugs

The (in)ability of the virus to replicate in the presence of antiretroviral drugs

Caused by changes in relevant part of the virus genome (mutations)

**The emergence of resistance is the inevitable consequence of incomplete suppression of viral replication (HIV/HBV/HCV) by the current antiretroviral drugs, and is a frequent and major limitation of antiviral therapy.**

# Viral escape

- The ability of a virus to escape antiviral pressure depends also upon the characteristics of drugs.
- Genetic barrier: The number of mutations required by the virus to develop a fully resistant virus.
  - **Low genetic barrier:** Drugs whose efficacy is lost with a single mutation.
  - **High genetic barrier:** Drugs whose efficacy is lost only after the sequential appearance and selection of a substantial number of mutations.

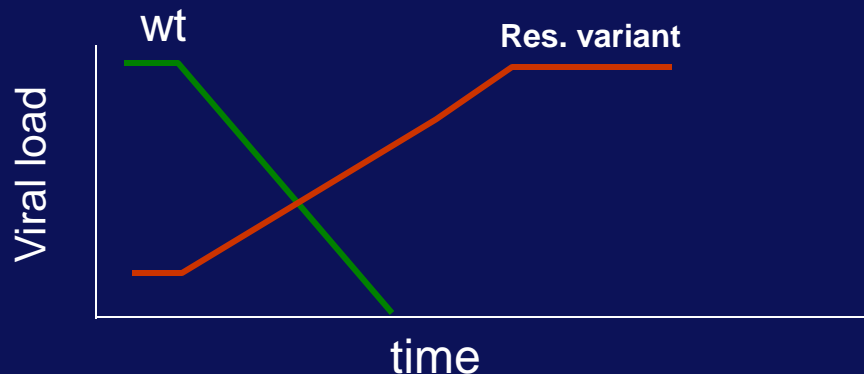
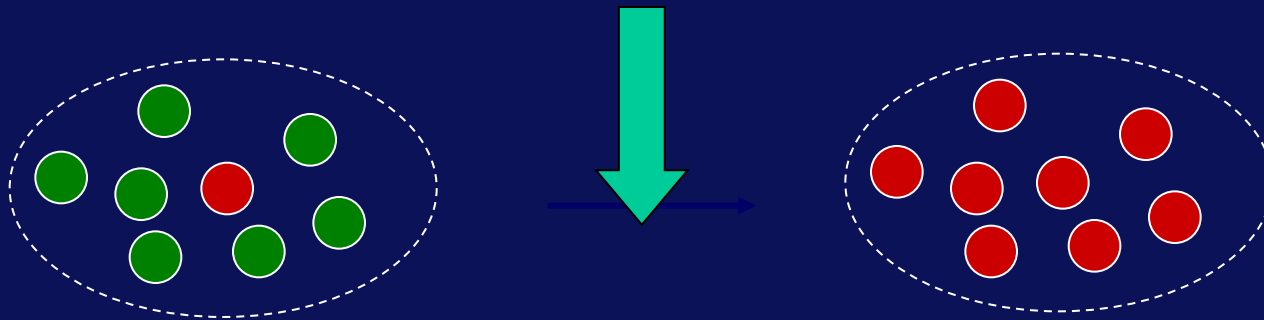


## Selection -> Resistance

A single mutation preexisting therapy, selected under antiviral pressure: an easy and *rapid phenomenon*

For instance NNRTI (K103N)  
(genetic barrier = 1)

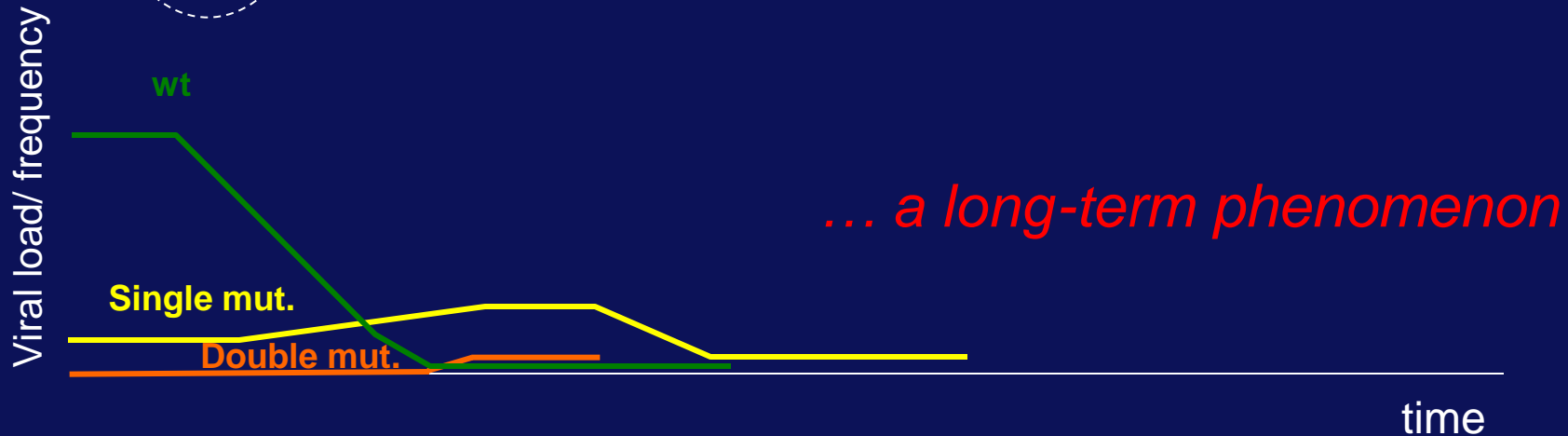
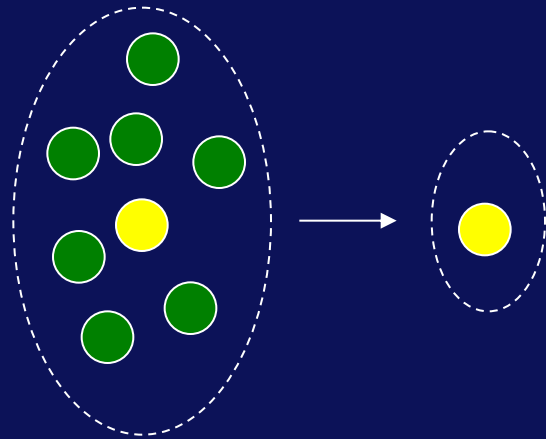
Inconsistent antiviral pressure



# Selection + Generation -> Resistance

A mutation preexisting therapy, conferring only marginal resistance, selected under antiviral pressure...

Selection



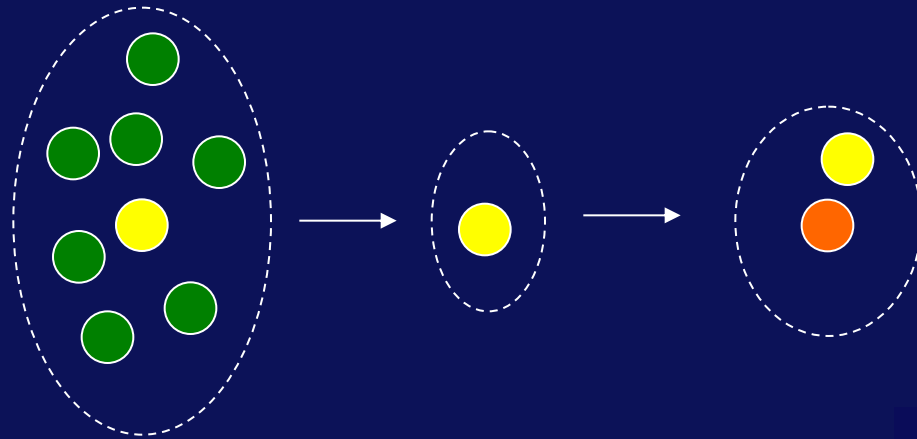
For instance, ritonavir-boosted PIs

## Selection + Evolution

A mutation preexisting therapy, conferring marginal resistance, selected under antiviral pressure, followed by the **generation of further mutations** while continuing antiviral therapy

Selection

Evolution toward high resistance and high fitness



Viral load/ frequency

wt

Single mut.

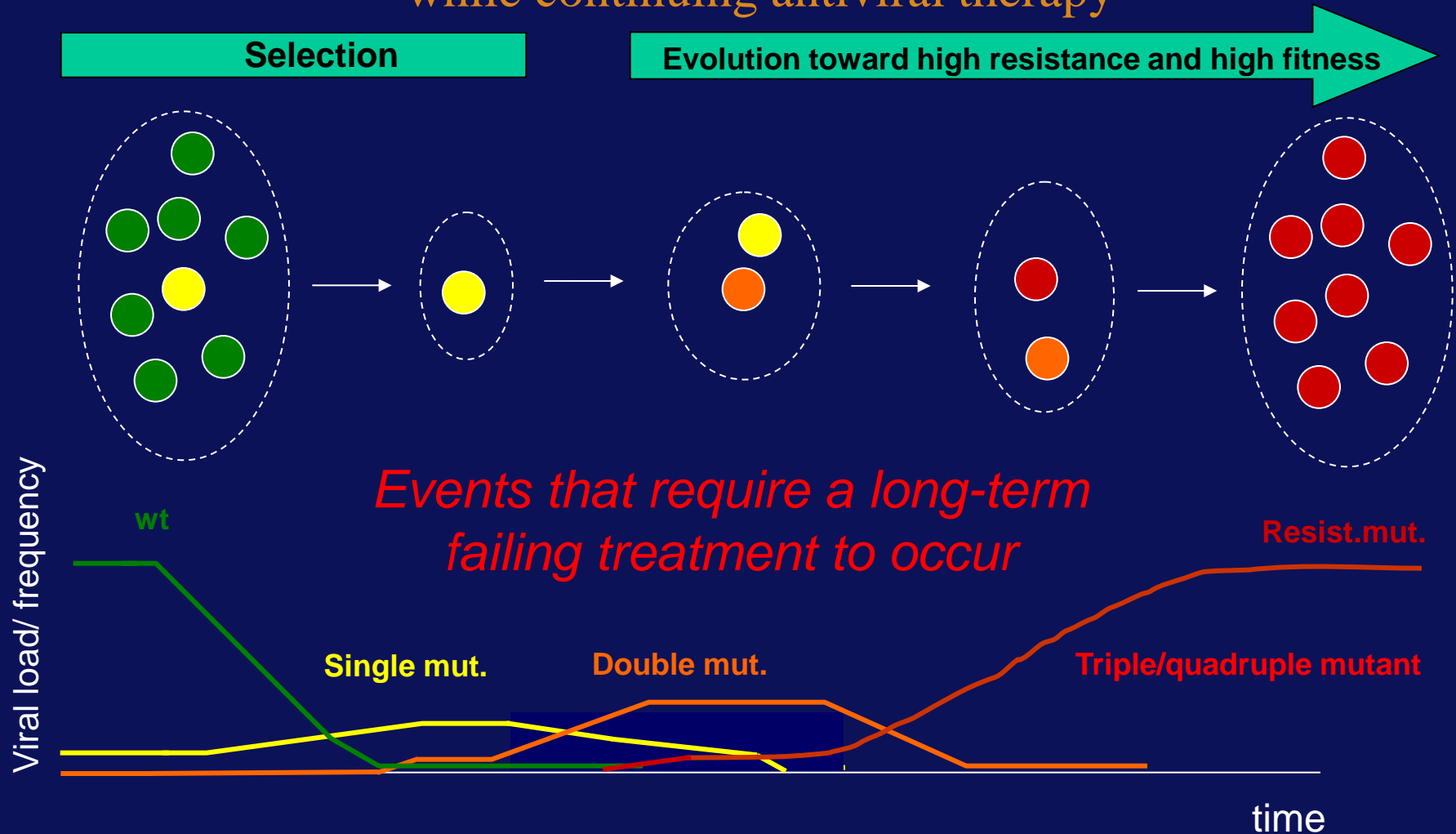
Double mut.

time

For instance, ritonavir-boosted PIs

## Selection + Generation -> Resistance

A mutation preexisting therapy, conferring marginal resistance, selected under antiviral pressure, followed by the generation of further mutations while continuing antiviral therapy



For instance boosted PI

**Genotypic resistance testing in HIV-1 infected patients is now recommended to guide the choice of antiretroviral therapy in clinical practice.**

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

**March 27, 2012**



Developed by the HHS Panel on Antiretroviral Guidelines for Adults and Adolescents – A Working Group of the Office of AIDS Research Advisory Council (OARAC)

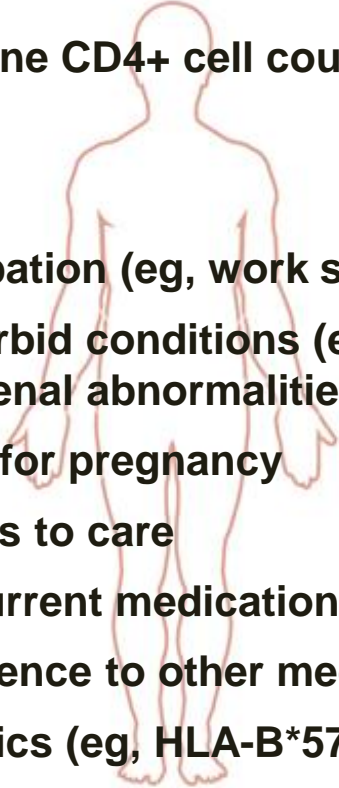
**Guidelines for the Clinical  
Management and Treatment  
of HIV Infected Adults in Europe  
European AIDS Clinical Society  
(EACS) *October 2011***

**Antiretroviral Treatment of Adult HIV Infection**  
2010 Recommendations of the International AIDS  
Society–USA Panel **JAMA®** *Thompson et al., July 2010*

# Considerations When Selecting First-line Antiretroviral Therapy

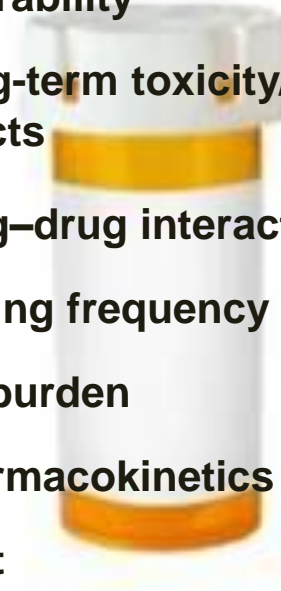
## Patient/Viral Factors

- Baseline HIV-1 RNA level
- Baseline CD4+ cell count
- Age
- Sex
- Occupation (eg, work schedule)
- Comorbid conditions (eg, CV risk, renal abnormalities)
- Plans for pregnancy
- Access to care
- Concurrent medications
- Adherence to other medications
- Genetics (eg, HLA-B\*5701)
- Viral tropism



## Antiretroviral Drug Factors

- Efficacy
- Baseline drug resistance
- Tolerability
- Long-term toxicity/metabolic effects
- Drug–drug interactions
- Dosing frequency
- Pill burden
- Pharmacokinetics
- Cost



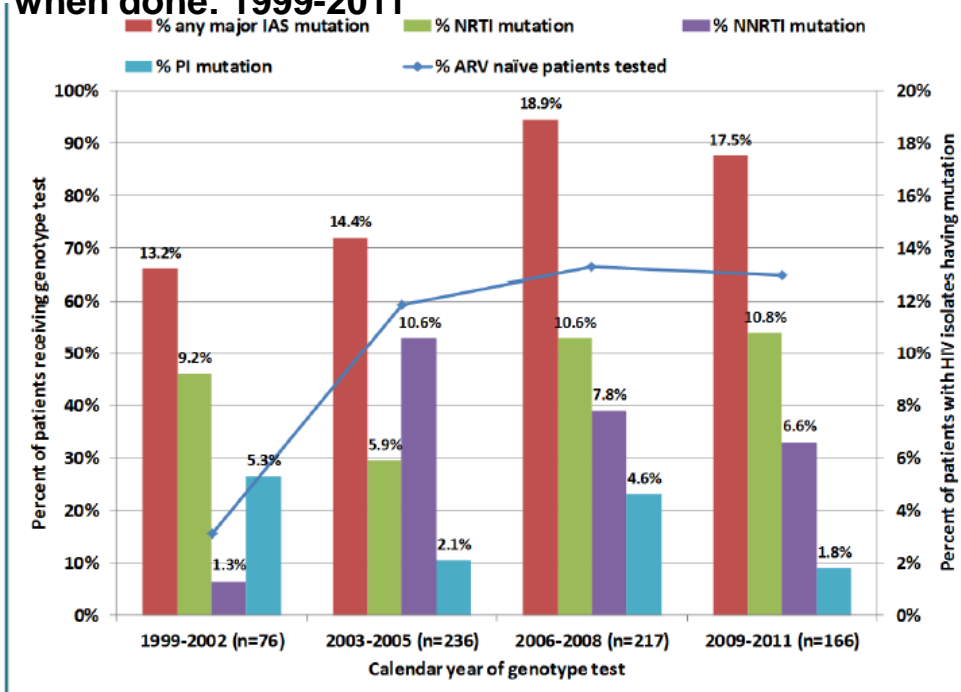


## Trends in Genotypic Resistance Testing Use and Results Among Antiretroviral-Naïve Patients in the HIV Outpatient Study (HOPS)

Kate Buchacz<sup>1</sup>, Benjamin Young<sup>2</sup>, Frank J Palella<sup>3</sup>, Carl Armon<sup>4</sup>, John T Brooks<sup>1</sup>, and the HIV Outpatient Study Investigators

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Percentage of ARV-naïve patients (total N= 1,484) having genotypic resistance test (GT), and prevalence of major IAS-USA resistance mutations when done: 1999-2011



No evidence of a statistically significant temporal increase in the frequency of any major IAS-USA mutation (13.2% in 1999-2002 to 17.5% in 2009-2011,  $p = 0.22$ ), or in the frequency of mutations to any of the three main drug classes was found.

## Original Article

# HIV-1 Subtypes and Primary Antiretroviral Resistance Mutations in Antiretroviral Therapy Naive HIV-1 Infected Individuals in Turkey

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<sup>2</sup>*Department of Infectious Diseases and Clinical Microbiology, Faculty of Medicine, University of Kocaeli, Kocaeli; and*

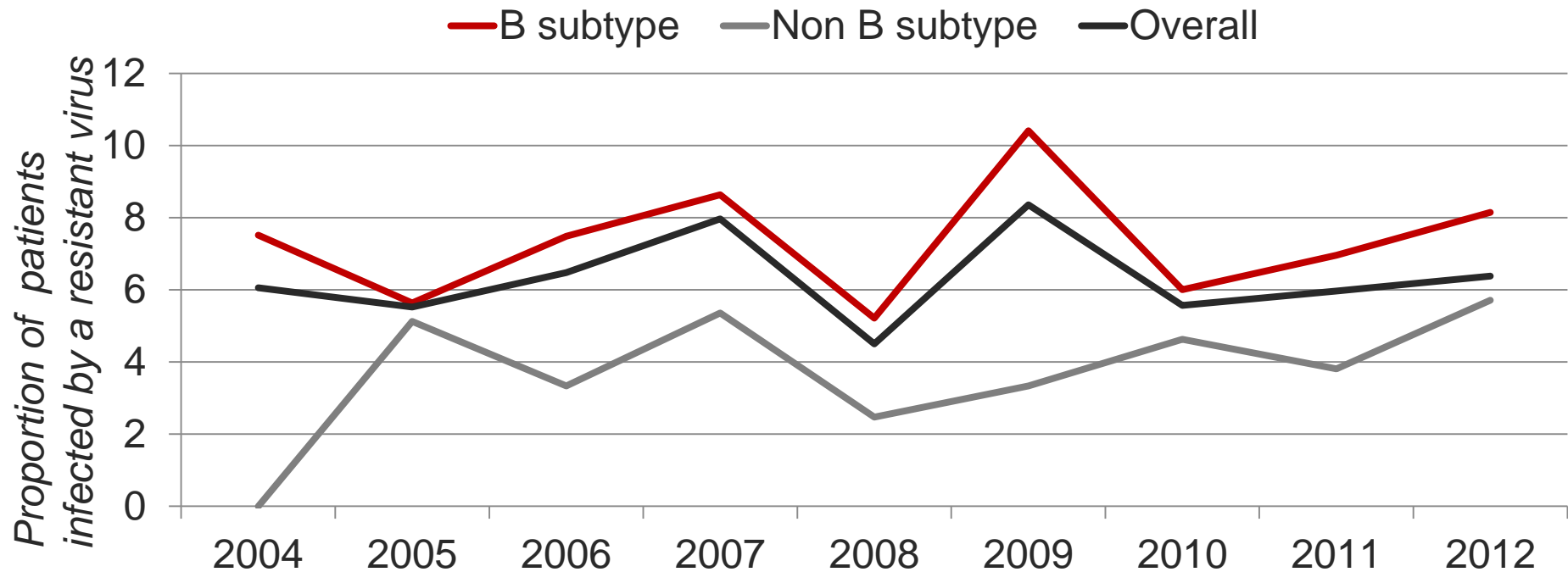
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(Received June 27, 2012. Accepted April 24, 2013)

**SUMMARY:** In this study, we determined the subtype distribution and the primary drug-resistant mutations in HIV-1 strains isolated from antiretroviral therapy (ART)-naive patients in Turkey. The study included 117 newly diagnosed HIV-1 positive Turkish patients. HIV-1 subtypes and circulating recombinant forms (CRFs) were identified by phylogenetic analysis (neighbor-joining method), and drug-resistant mutations were analyzed according to the 2009 World Health Organization list of surveillance drug-resistant mutations. Subtype CRFs (CRF 02\_AG, CRF 01\_AE, CRF 12\_BF and CRF 03\_AB; 47%, 55/117) and B (33.3%, 39/117) were identified as the most common occurring HIV-1 subtypes in Turkey. The patients had primary antiretroviral resistance mutations to nucleos(t)ide reverse transcriptase (RT) inhibitors (NRTIs) (M41L, T215C, T215D, and K219Q), non-nucleoside RT inhibitors (NNRTIs; K103N), and protease inhibitors (PIs; I47V, G73S). The prevalence of overall primary antiretroviral resistance was 7.6% (9/117) in HIV-1 patients from Turkey and drug-resistant rate for NRTIs, NNRTIs, and PIs were 4.2% (5/117), 1.7% (2/117), and 1.7% (2/117), respectively. In this study, various CRFs of HIV-1 were determined, for the first time, in Turkey. The prevalence of HIV-1 primary drug-resistant mutations in ART-naive patients suggested that resistance testing should be incorporated as an integral part of HIV management, and the choice of a first-line therapy regime should be guided by the results of genotypic resistance in Turkey.



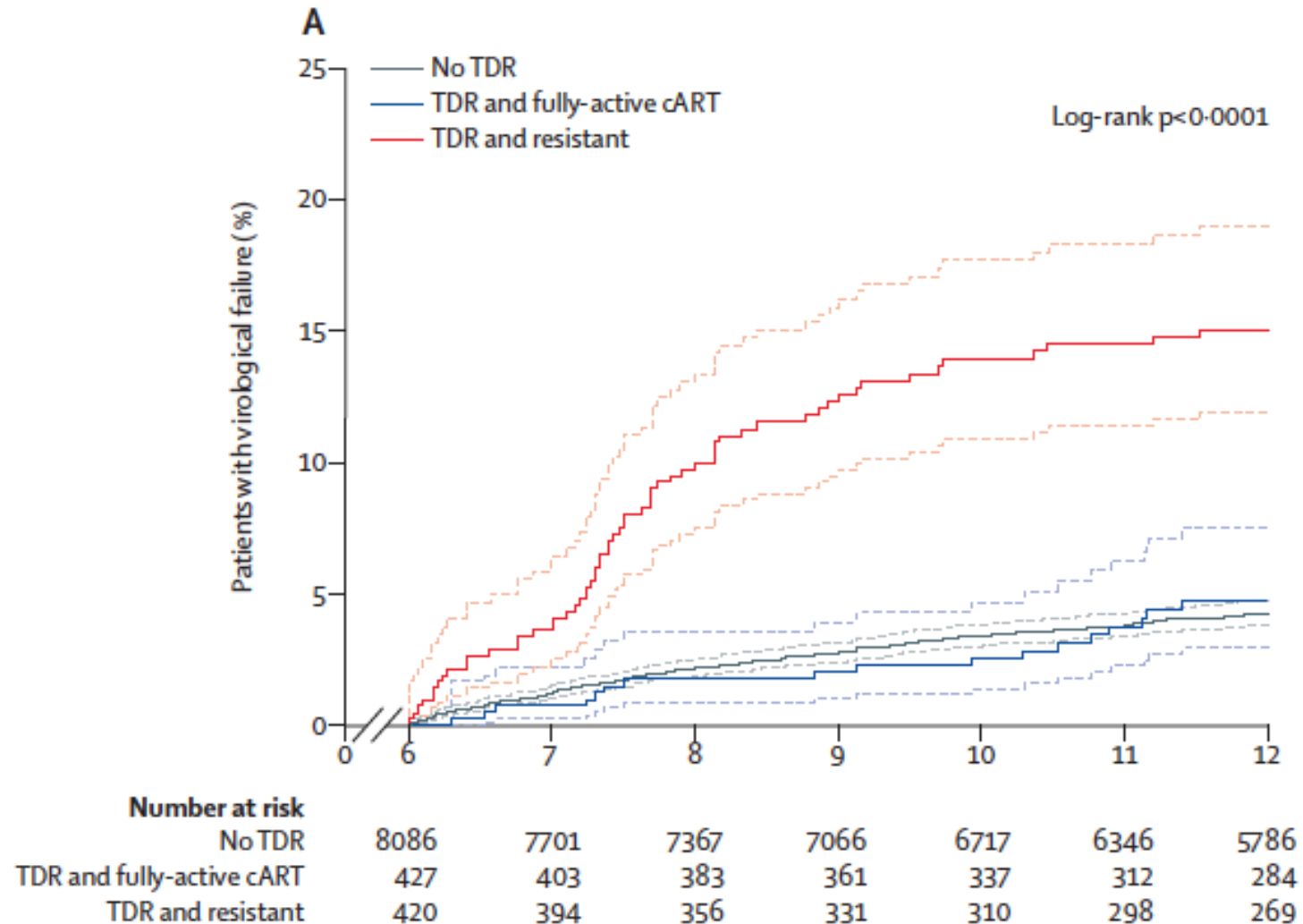
# ***Prevalence of transmitted drug resistance over the years in Central Italy***



*The prevalence of TDR in non-B infected patients was constantly lower than that observed in B infected patients*

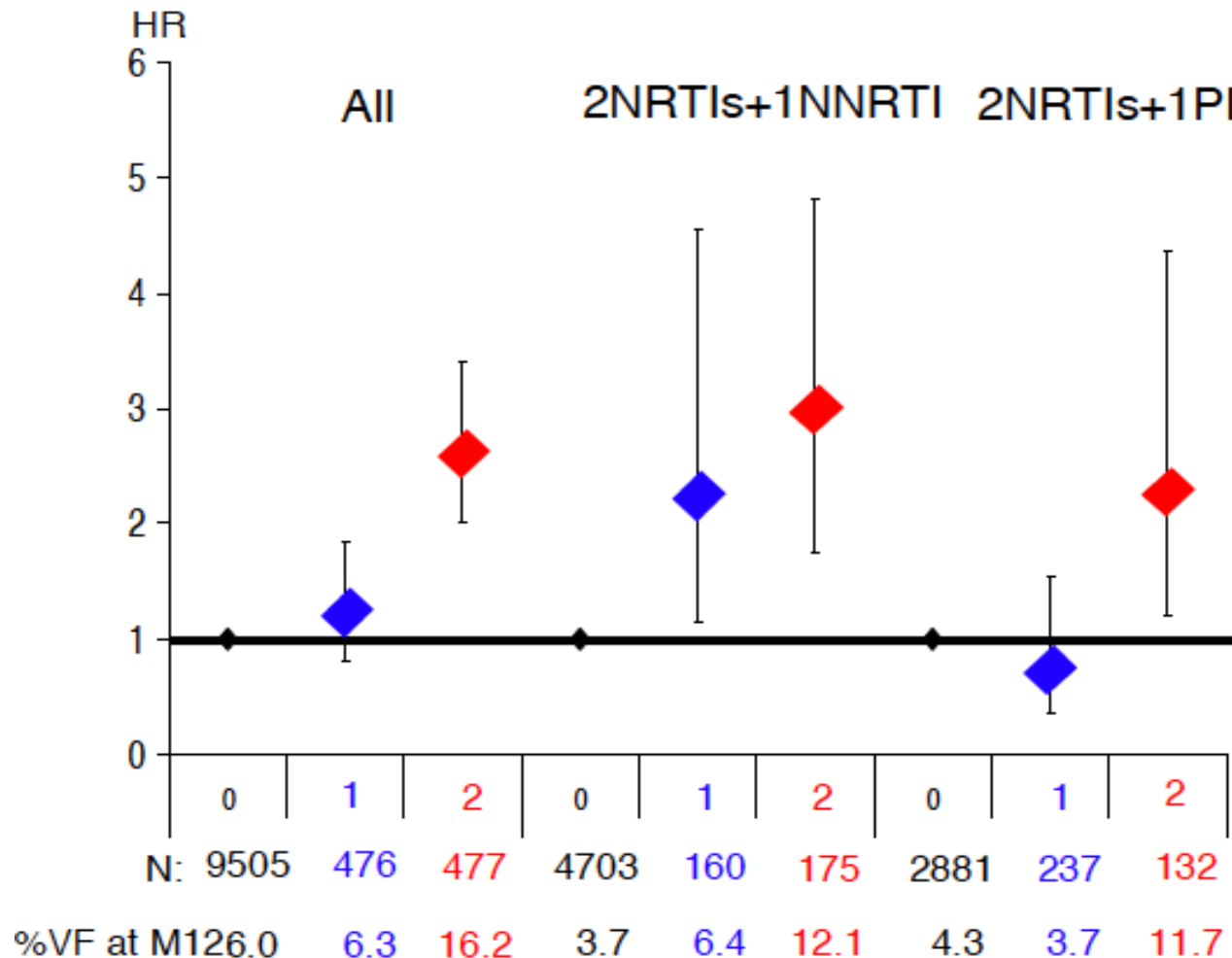
**Analysis on the Sendih cohort (N=2464)**

# Transmitted drug resistance is associated with a poorer virological response when patients received cART containing $\geq 1$ drug not fully active



VF rates at M12 were 6.0% (95% confidence interval [CI]: 5.5; 6.5), 6.3% (4.2; 9.3) and 16.2% (13.0; 20.1) for no TDR group, TDR and fully active group and TDR and resistant group, respectively.

# Virological failure rates at M12 (Kaplan Meier estimates) are higher when NNRTI-based regimens are used in patients carrying drug-resistant viruses at baseline



Amongst patients receiving 2 NRTIs + 1 NNRTI, **those harbouring  $\geq 1$  mutation but predicted to receive a fully active cART had a higher risk for VF** (HR: 2.3, 95% CI: 1.2; 4.6,  $P=0.02$ ) **than those carrying no mutation.**

**$\geq 1$  mutation but receiving a treatment predicted to be fully active**

**$\geq 1$  mutation and at least low-level resistance to  $\geq 1$  prescribed drug**

# Emergence of Drug Resistance in HIV Type 1–Infected Patients after Receipt of First-Line Highly Active Antiretroviral Therapy: A Systematic Review of Clinical Trials

**Background.** Resistance to antiretroviral combination therapy is associated with increased mortality. Understanding the relative risks of emerging resistance to first-line therapy is of importance for both resource-rich and resource-poor settings.

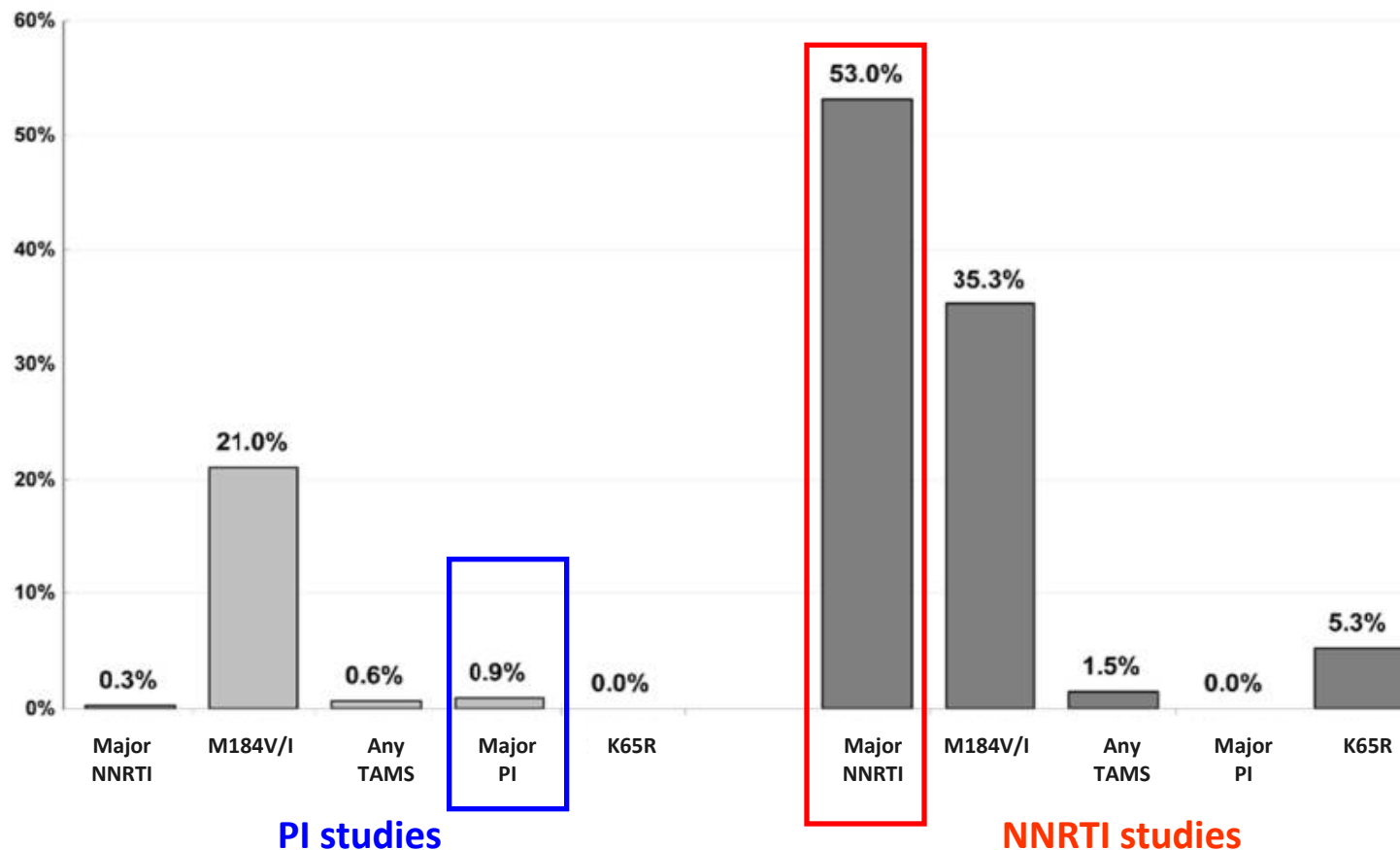
**Methods.** We undertook an overview of clinical trials of adults receiving first-line highly active antiretroviral therapy (HAART), which consisted of dual nucleoside reverse-transcriptase inhibitors (NRTIs) combined with a third agent (either a nonnucleoside reverse-transcriptase inhibitor [NNRTI] or a ritonavir-boosted protease inhibitor [bPI]). The primary outcome measures were incidences of mutations conferring resistance to key drugs (NRTIs, NNRTIs, or bPIs) per trial at week 48. For meta-analysis, inverse-variance weighting was used to create estimates of overall incidences per group, with exact 95% confidence intervals (95% CIs).

**Results.** The study included 20 clinical trials that comprised 30 treatment arms and 7970 patients. Virologic failure at 48 weeks occurred in 4.9% (95% CI, 3.9%–6.1%) of NNRTI recipients, compared with 5.3% (95% CI, 4.4%–6.4%;  $P = .50$ ) of bPI recipients. Of failures that were successfully genotyped, the M184V mutation in the HIV reverse transcriptase (lamivudine resistance) occurred in 35.3% (95% CI, 29.3%–41.6%) of patients who started NNRTI-based HAART, compared with 21.0% (95% CI, 14.4%–28.8%;  $P < .001$ ) for those who received a bPI. For the K65R mutation in the HIV reverse transcriptase (multinucleoside resistance), incidences were 5.3% (95% CI, 2.4%–9.9%) and 0.0% (95% CI, 0.0%–3.6%;  $P = .01$ ), respectively, in patients treated with non-zidovudine-containing regimens. Resistance to the third agent (an NNRTI or PI) occurred in 53% (95% CI, 46%–60%) and 0.9% (95% CI, 0.0%–6.2%;  $P < .001$ ) of such patients, respectively.

**Conclusions.** Initial therapy with bPI-based regimens resulted in less resistance within and across drug classes. This finding is of particular significance for the developing world, where rates of resistance to NRTIs and NNRTIs at 48 weeks are much higher than has been seen in both cohorts and clinical trials in well-resourced countries.

*Gupta et al., Clinical Infectious Diseases 2008; 47:712–22*

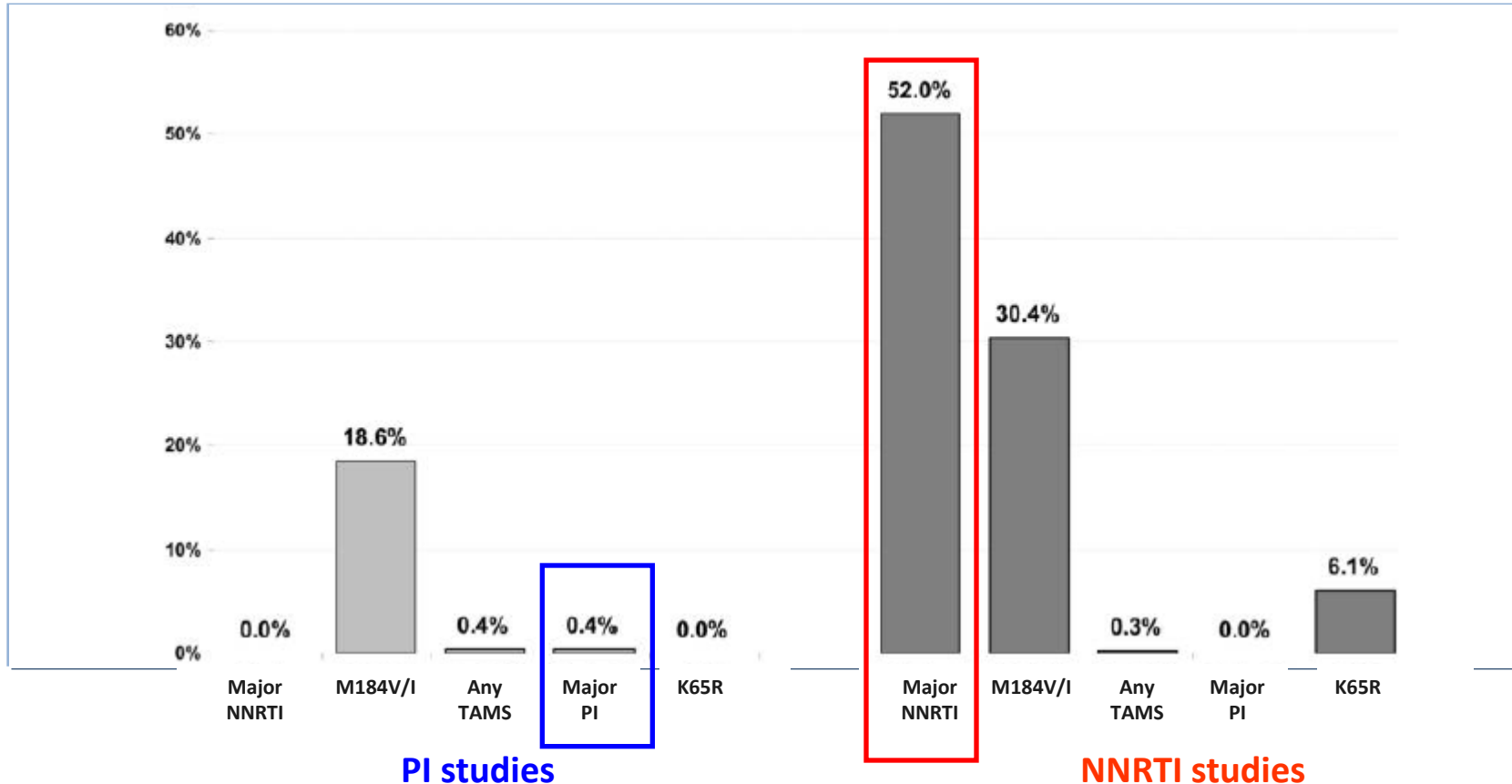
The incidence of resistance to the third agent (an **NNRTI** or **PI**) was significantly lower in patients starting a regimen containing a **ritonavir-boosted PI** ( $P < 0.001$ ) compared that in patients starting an **NNRTI**-based HAART at virologic failure at **48 wks**



Week 48 incidence of genotypic resistance at virologic failure. Genotype analysis.

*Gupta et al., Clinical Infectious Diseases 2008*

Similar results were obtained at 96 wks of treatment



Week 96 incidence of genotypic resistance at virologic failure. Genotype analysis.

*Gupta et al., Clinical Infectious Diseases 2008*



# Emergence of HIV-1 Drug Resistance in Previously Untreated Patients Initiating Combination Antiretroviral Treatment

## *A Comparison of Different Regimen Types*

Viktor von Wyl, MSc; Sabine Yerly, MSc; Jürg Böni, DVM; Philippe Bargisser, PhD; Thomas Klimkait, PhD; Manuel Battegay, MD; Hansjakob Furrer, MD; Amalio Telenti, MD; Bernard Hirschel, MD; Pietro L. Vernazza, MD; Enos Bernasconi, MD; Martin Rickenbach, MD; Luc Perrin, MD; Bruno Ledergerber, PhD; Huldrych F. Günthard, MD; for the Swiss HIV Cohort Study

**Background:** Standard first-line combination antiretroviral treatment (cART) against human immunodeficiency virus 1 (HIV-1) contains either a nonnucleoside reverse transcriptase inhibitor (NNRTI) or a ritonavir-boosted protease inhibitor (PI/r). Differences between these regimen types in the extent of the emergence of drug resistance on virological failure and the implications for further treatment options have rarely been assessed.

**Methods:** We investigated virological outcomes in patients from the Swiss HIV Cohort Study initiating cART between January 1, 1999, and December 31, 2003, with an unboosted PI, a PI/r, or an NNRTI and compared genotypic drug resistance patterns among these groups at treatment failure.

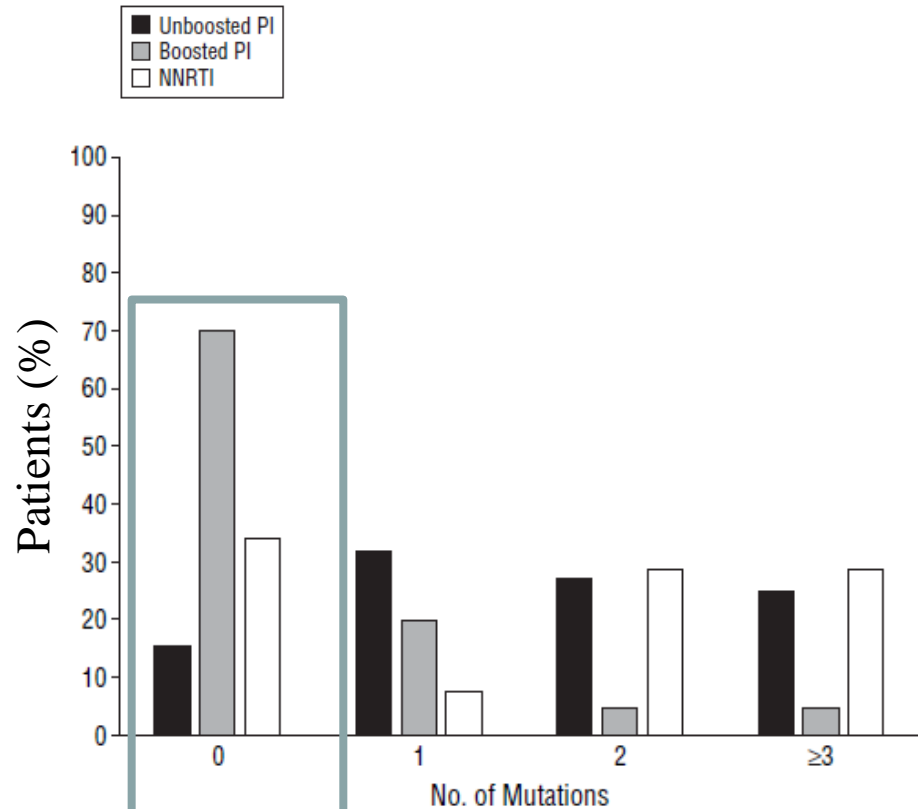
**Results:** A total of 489 patients started cART with a PI, 318 with a PI/r, and 803 with an NNRTI. A total of 177 virological failures were observed (108 [22%] PI failures, 24 [5%] PI/r failures, and 45 [6%] NNRTI failures). The failure rate was highest in the PI group (10.3 per 100 person-years; 95% confidence interval [CI], 8.5-

12.4). No difference was seen between patients taking a PI/r (2.7; 95% CI, 1.8-4.0) and those taking an NNRTI (2.4; 95% CI, 1.8-3.3). Genotypic test results were available for 142 (80%) of the patients with a virological treatment failure. Resistance mutations were found in 84% (95% CI, 75%-92%) of patients taking a PI, 30% (95% CI, 12%-54%) of patients taking a PI/r, and 66% (95% CI, 49%-80%) of patients taking an NNRTI ( $P < .001$ ). Multidrug resistance occurred almost exclusively as resistance against lamivudine-emtricitabine and the group-specific third drug and was observed in 17% (95% CI, 9%-26%) of patients taking a PI, 10% (95% CI, 0.1%-32%) of patients taking a PI/r, and 30% (95% CI, 33%-67%) of patients taking an NNRTI ( $P < .001$ ).

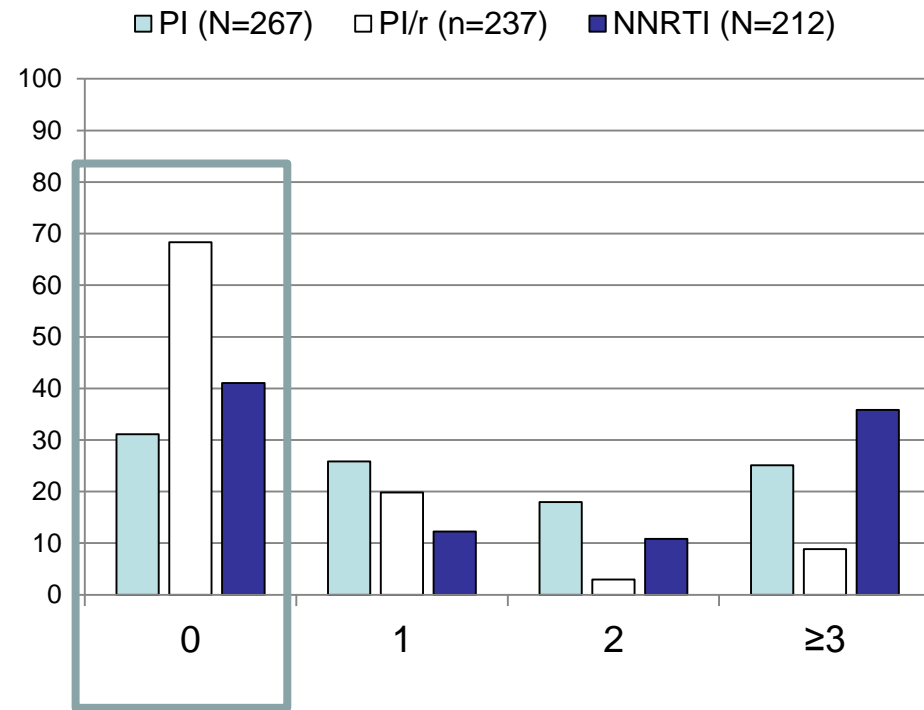
**Conclusions:** Regimens that contained a PI/r or an NNRTI exhibited similar potency as first-line regimens. However, the use of a PI/r led to less resistance in case of virological failure, preserving more drug options for the future.

*Arch Intern Med.* 2007;167(16):1782-1790

# Number of International AIDS Society–USA major mutations detected in patients failing the first line regimen



Von Wyl et al., Arch Intern Med 2009



Santoro et al.. Unpublished data

Number of mutations per drug class



# The prevalence of resistance mutations varied according to the viral load.

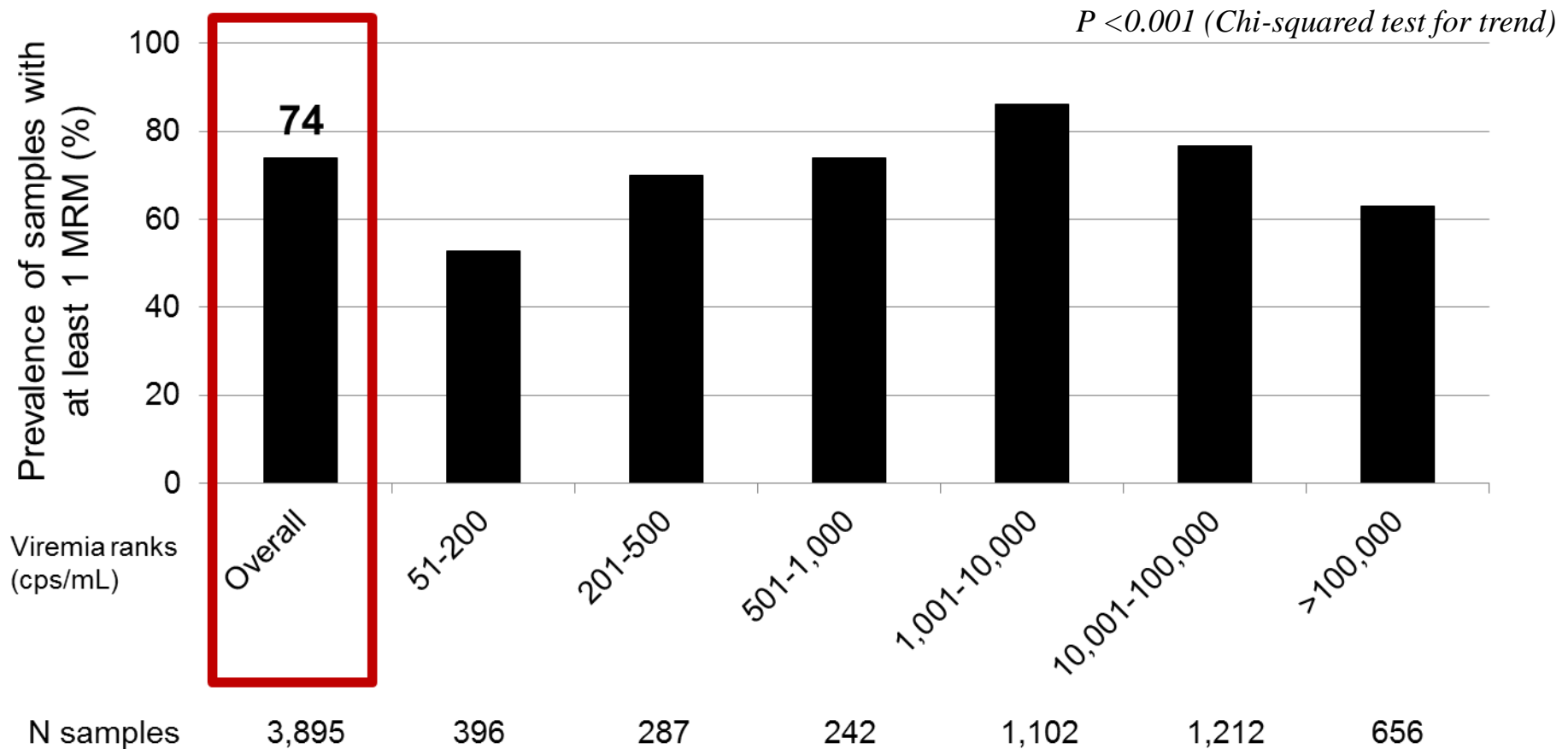
Detection of at least 1 resistance mutation, both overall and specific to 1 drug class, was **most frequent** among tests performed at viral loads of **300–10,000 copies/mL** and decreased as the viral load increased above these levels.

**Table 2. Prevalence and Relative Risk (RR) of Detection of Drug Resistance According to Viral Load**

Viral load, copies/mL	All patients			Patients receiving NRTIs			Patients receiving NNRTIs			Patients receiving PIs		
	<i>n</i>	Any resistance mutation	RR (95% CI)	<i>n</i>	NRTI resistance mutation	RR (95% CI)	<i>n</i>	NNRTI resistance mutation	RR (95% CI)	<i>n</i>	PI resistance mutation	RR (95% CI)
<300	449	270 (60)	0.94 (0.87–1.01)	410	219 (53)	0.89 (0.81–0.98)	126	61 (48)	0.85 (0.72–1.00)	193	55 (29)	0.85 (0.70–1.04)
300–999	552	399 (72)	0.99 (0.94–1.04)	508	345 (68)	1.01 (0.94–1.07)	161	124 (77)	1.04 (0.94–1.15)	237	90 (38)	1.00 (0.87–1.16)
1000–2999	1119	851 (76)	1	994	712 (72)	1	315	247 (78)	1	423	182 (43)	1
3000–9999	1311	1014 (77)	1.01 (0.97–1.05)	1135	850 (75)	1.01 (0.96–1.06)	363	281 (77)	1.01 (0.94–1.09)	483	224 (46)	0.89 (0.79–1.01)
10,000–29,999	1323	892 (67)	0.91 (0.87–0.95)	1003	700 (70)	0.92 (0.88–0.97)	298	220 (74)	0.96 (0.88–1.04)	454	199 (44)	0.87 (0.77–0.99)
30,000–99,999	1433	862 (60)	0.84 (0.80–0.88)	1014	595 (59)	0.79 (0.74–0.84)	282	170 (60)	0.82 (0.74–0.92)	462	192 (42)	0.85 (0.75–0.96)
≥100,000	1674	811 (48)	0.69 (0.65–0.74)	1076	482 (45)	0.61 (0.57–0.66)	319	155 (49)	0.67 (0.59–0.76)	507	157 (31)	0.64 (0.55–0.75)

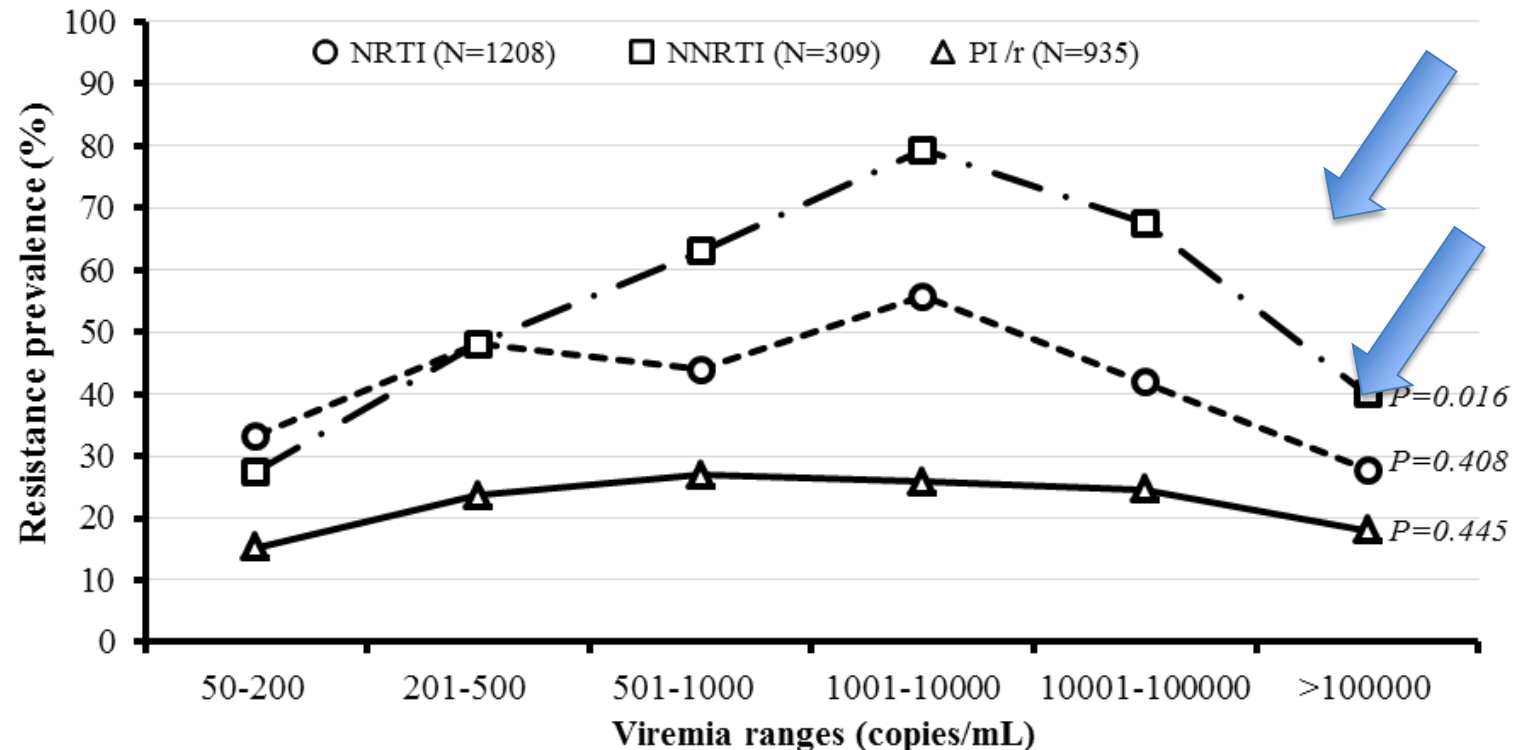
**NOTE.** Data are no. (%) of drug resistance tests, unless otherwise indicated, following adjustment for the variables shown in Table 1. CI, confidence interval; NNRTI, nonnucleoside reverse-transcriptase inhibitor; NRTI, nucleoside reverse-transcriptase inhibitor; PI, protease inhibitor.

**The overall prevalence of samples with at least one major resistance mutation (MRM) in 3,895 genotypic resistance tests from 2,200 treated-patients with viremia>50 copies/mL is 74%.**



**Prevalence of samples with at least one major resistance mutation in patients failing PIs (boosted or unboosted), NRTIs or NNRTIs, stratified by viremia.**

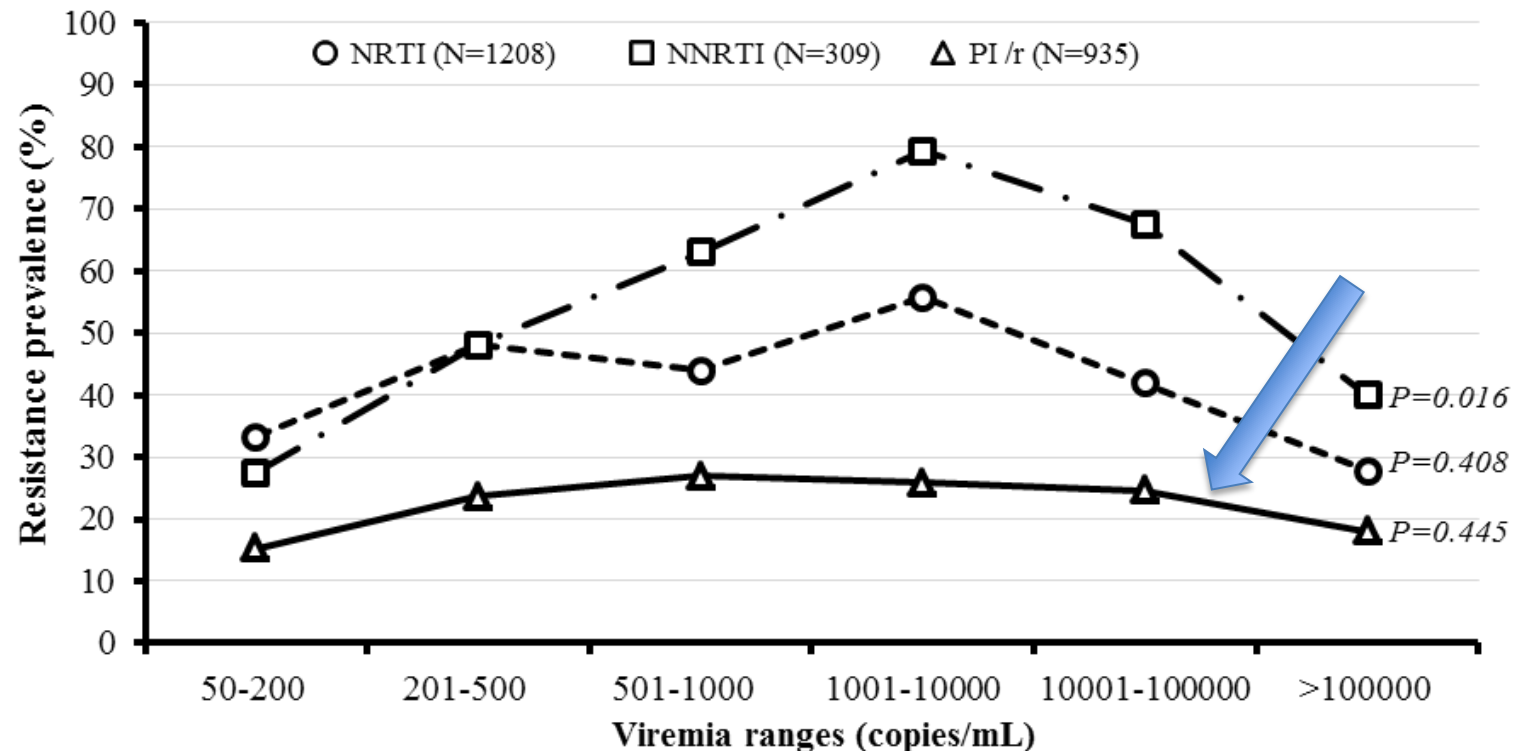
By analyzing samples genotyped from patients failing their treatment between 2008-2012, the resistance to NRTI and NNRTI varied according to viral load strata, with a still considerable prevalence of resistance in samples with viremia levels  $\leq 1000$  copies/mL.



NRTI	196	123	82	283	315	209
NNRTI	40	27	27	87	83	45
PI/r	150	106	63	209	240	167

Resistance to NRTI, NNRTI or PI/r classes in samples collected from January 2008 to December 2012 stratified for plasma viremia ranges

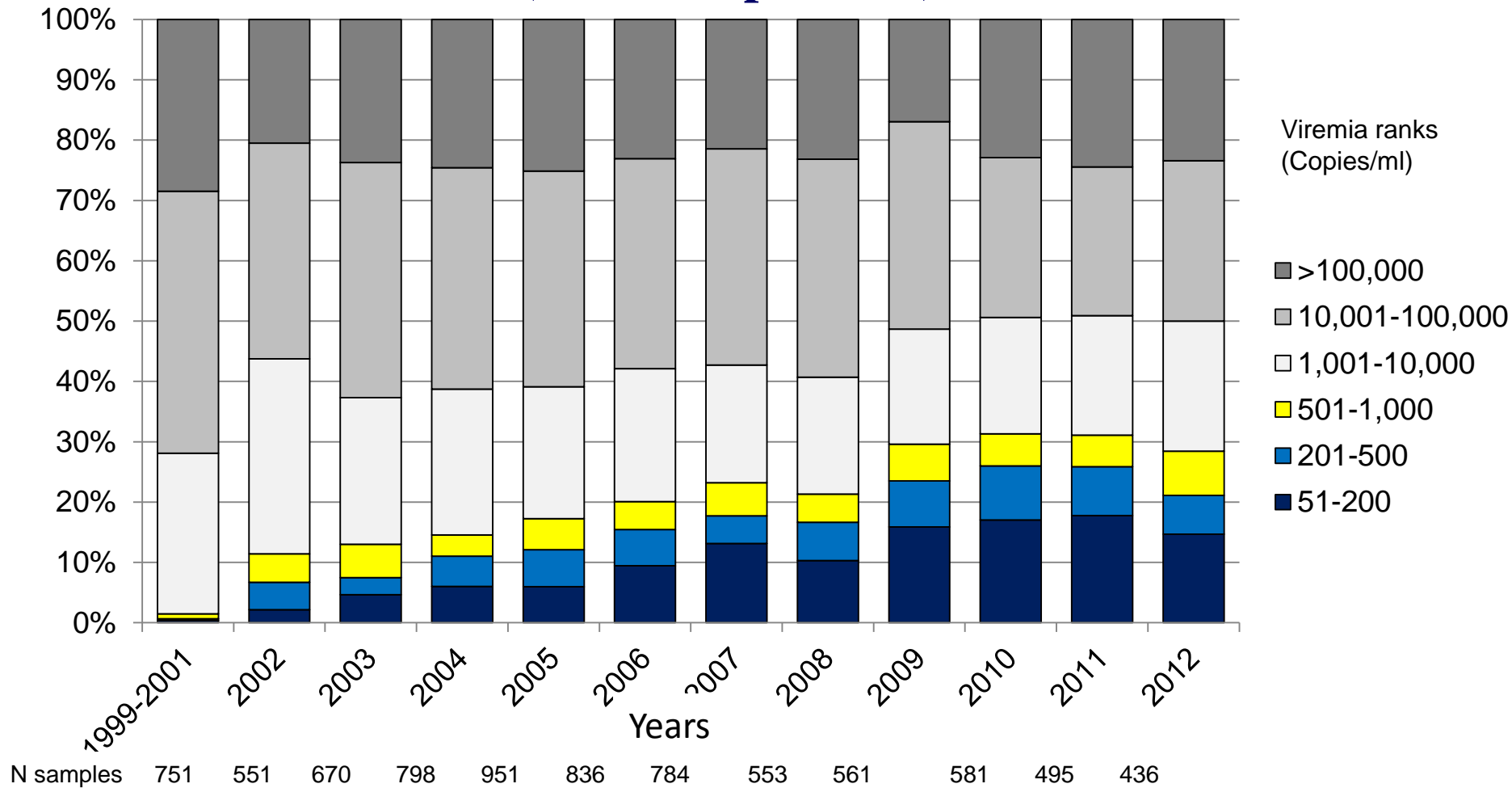
By contrast, the prevalence of PI-resistance was not influenced by viral load strata because it was very limited among all failures and was almost zero in patients failing their first line PI/r containing regimen.



NRTI	196	123	82	283	315	209
NNRTI	40	27	27	87	83	45
PI/r	150	106	63	209	240	167

Resistance to NRTI, NNRTI or PI/r classes in samples collected from January 2008 to December 2012 stratified for plasma viremia ranges

# In the last years an increased number of genotypic resistance tests were requested for patients failing with low viremia values (50-500 copies/mL).



**7,967 genotypic requests from plasma samples of treatment experienced patients stratified by viremia and years.**

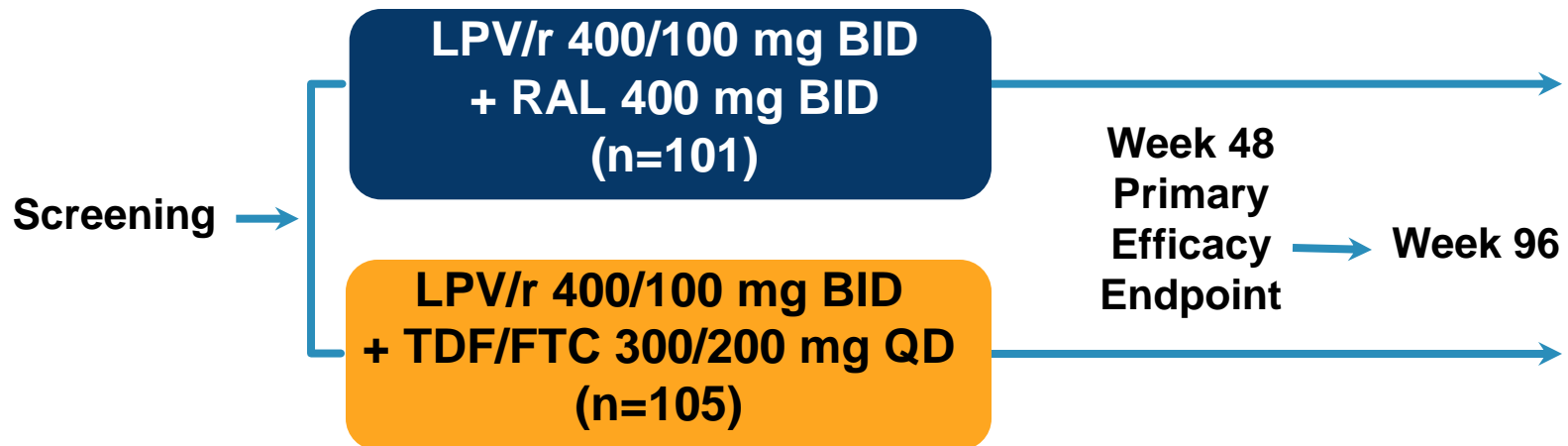
Drugs with high genetic barrier decrease  
the risk of development of resistance,  
and also reduce the resistance of  
companion drugs

This preserves future therapeutic  
options!!

# LPV/r + RAL vs. LPV/r + TDF/FTC in Treatment-Naive Subjects: PROGRESS Study Design

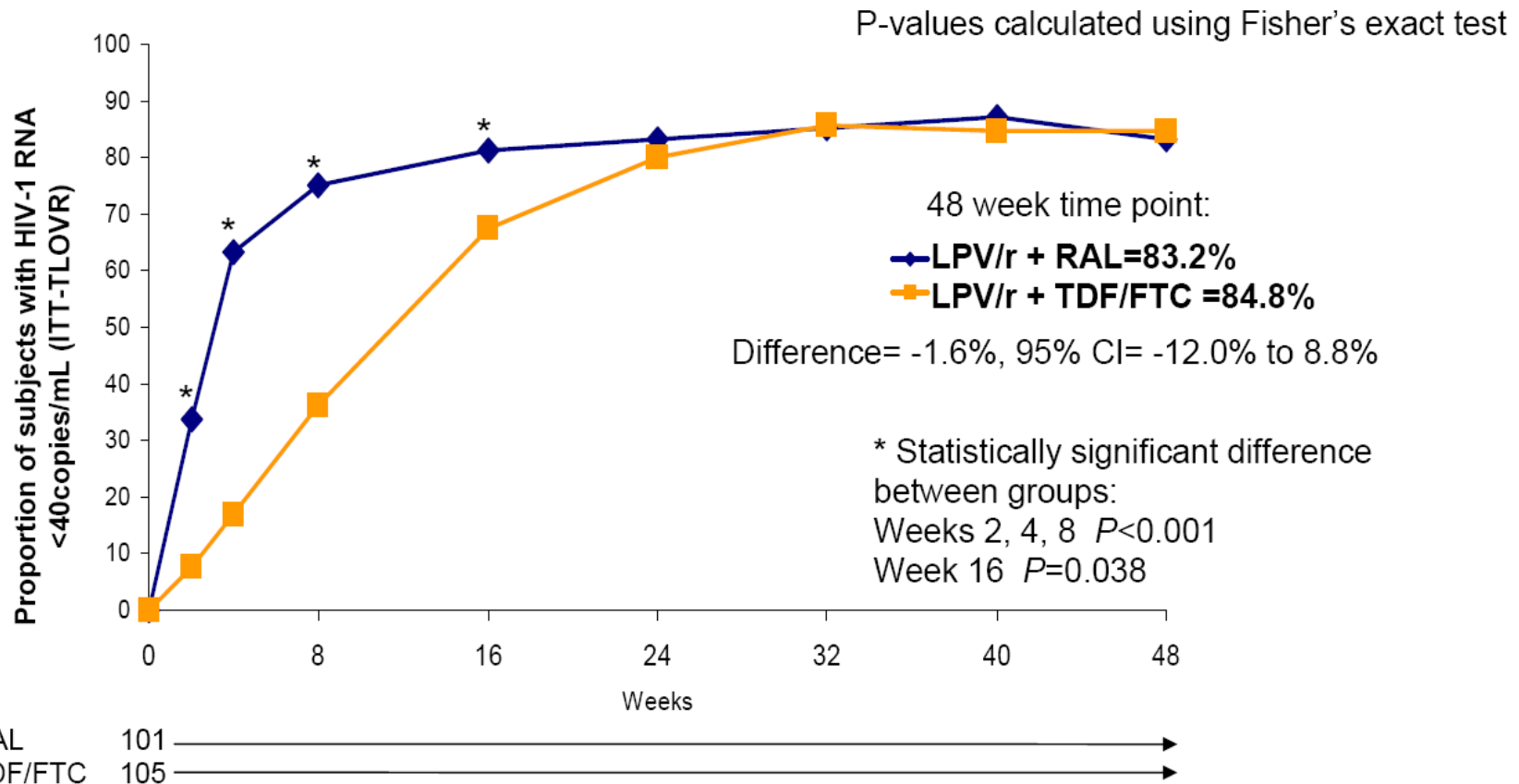
## Inclusion Criteria for PROGRESS (M10-336)

- HIV-1 infection
- ARV-naïve
- Plasma HIV-1 RNA >1000 copies/mL
- Any CD4<sup>+</sup> T-cell count



- Primary endpoint: plasma HIV-1 RNA <40 copies/mL at week 48 (FDA ITT TLOVR)
- Non-inferiority assessed by 95% CI for the difference ([LPV/r + RAL] – [LPV/r + TDF/FTC]) using a -20% threshold
- If non-inferiority with respect to the -20% margin was demonstrated, then non-inferiority with respect to a -12% margin was to be evaluated

# Primary Efficacy Endpoint at Week 48: Proportion of Subjects Responding (FDA ITT TLOVR)



**LPV/r + RAL was non-inferior to LPV/r + TDF/FTC in treatment-naïve subjects at 48 weeks**



# Emergence of Resistance Mutations\*

- **7 subjects (4 LPV/r + RAL and 3 LPV/r + TDF/FTC) met the protocol-defined criteria for resistance testing**
  - N155H mutation detected in 1 LPV/r + RAL subject
  - M184V mutation detected in 1 LPV/r + TDF/FTC subject

Study Drug	Number of subjects with new mutations	
	LPV/r + RAL	LPV/r + TDF/FTC
LPV	0/4	0/3
RAL †	1/3	N/A
TDF	0/4	0/3
FTC	0/4	1/3

**\*LPV-associated mutations:** Major: V32I, I47V/A, L76V, V82A/F/T/S. Minor: L10F/I/R/V, K20M/R, L24I, L33F, M46I/L, I50V, F53L, I54/V/L/A/M/T/S, L63P, A71V/T, G73S, I84V, or L90M

**RAL-associated mutations:** Y143R/H/C, Q148H/K/R, or N155H

**TDF-associated mutations:** K65R or K70E

**FTC-associated mutations:** K65R or M184V/I

† 1 RAL sample not available for testing after meeting protocol-defined criteria

N/A-not applicable

# Conclusions

**The characteristics of HIV infection have deeply changed, as well as the expectations of antiviral therapy**

**The construction of antiretroviral therapy must be designed taking into account the long-term strategy, and not the mere control of short term viral replication.**

**The design of regimens able to control the virus over a long-time period must take into account genetic barrier and potency, since the first regimen**

**In this frame, selection of the best therapy, based also on resistance testing, warrants the best result for each single patient**