Clinical Case – HIV Resistance

Santoro Maria

Università di Roma Tor Vergata

HIV CLINICAL TOPICS

Update on Antiretroviral Therapy Workshop

THURSDAY, 19TH AND FRIDAY, 20TH SEPTEMBER 2024

Recinte Modernista de Sant Pau, Barcelona

Disclosures

Santoro Maria has received funds for attending symposia, speaking and organizing educational activities from

- *MSD*
- *ViiV Healthcare*
- *Gilead Sciences*

Italian man

- **Italian man (81 years),** Caucasian, 167cm height, 80kg weight, ex-smoker (from 20yo to 32yo), no drug-user
- **HIV Diagnosis:** 1998
- **Risk factor:** Homosexual
- **CDC Classification:** C3 (Wasting syndrome)
- **No co-infection with HBV/HCV**
- **Previous syphilis**
- **Previous tuberculosis**
- **Current Comorbidities:** hypercholesterolemia, hypertryglicerides, chronic renal failure, hypovitaminosis D, benign prostatic hyperplasia, haemorrhoidal varicles
- Previously followed in another center

- ***ARRIVED AT OUR ATTENTION in September 2015***

Italian man

- **Non-ART treatments:** Atorvastatin, Omega-3, Vitreoxygen, Dutasteride, Tamsulosin, Cholecalciferol, Zofenopril, Barnidipin
- **cART history:**

1. EFV/TDF/FTC (undefined period from 2000?, autonomously suspended)

Italian man

HIV-RNA= 972,437 copies/mL DATE: 23/09/2015
CD4=66 cells/mm^3 (4%)

GRT ON PLASMA HIV RNA (Sanger)

KNOWN MUTATIONS*

- **PROTEASE (PR): M36I, L63P, I93L**
- **REVERSE TRANSCRIPTASE (RT): V179D/V**
- **INTEGRASE (IN): NONE**

RESULT:
PR: Presence of 3 secondary mutations
RT: Presence of 1 mutation
IN: Absence of mutations conferring resistance to Integrase inhibitors

TROPISM GP120/V3: Virus with dual tropism X4/R5 (FPR: 7.1%)

**http://hivdb.stanford.edu/*

POTENTIALLY EFFECTIVE DRUGS:
PR: All
RT: All (see interpretation)
IN: All
CCR5: None (see interpretation)

NOTE: Phylogenetic analysis of the nucleotide sequence identified a B subtype of HIV-1. Test performed with Sanger sequencing.

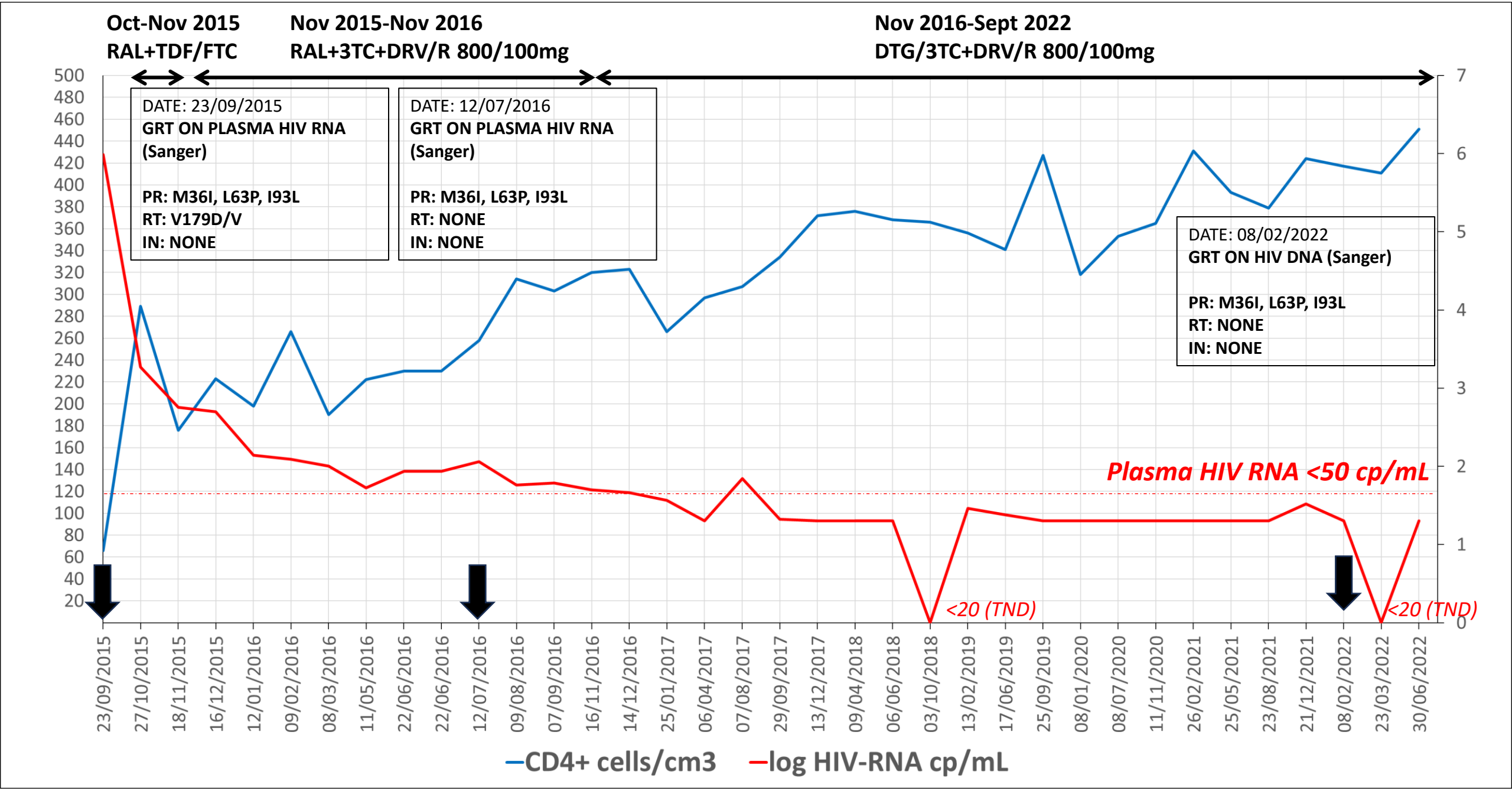
VIROLOGICAL INTERPRETATION: In the context of a wild type picture, the presence of the V179D mutation in Reverse Transcriptase is reported, which is able to modulate the efficacy of NNRTIs. It is possible that this mutation is derived from a previous failure to the three-drug regimen reported in the anamnesis. The very high viremia and the compromised immunological picture suggest not to consider the NNRTI class as a first choice: however, its use is possible at the time of therapeutic switches with stably undetectable viremia, in the context of high adherence.
The tropism test indicates the presence of a dual tropic strain (with relative prevalence R5), in which maraviroc should not be considered. It could maintain an efficacy (however partial) only in the context of therapy with other effective drugs. It does not currently represent a first choice. It is appropriate to maintain an adequately high genetic barrier.

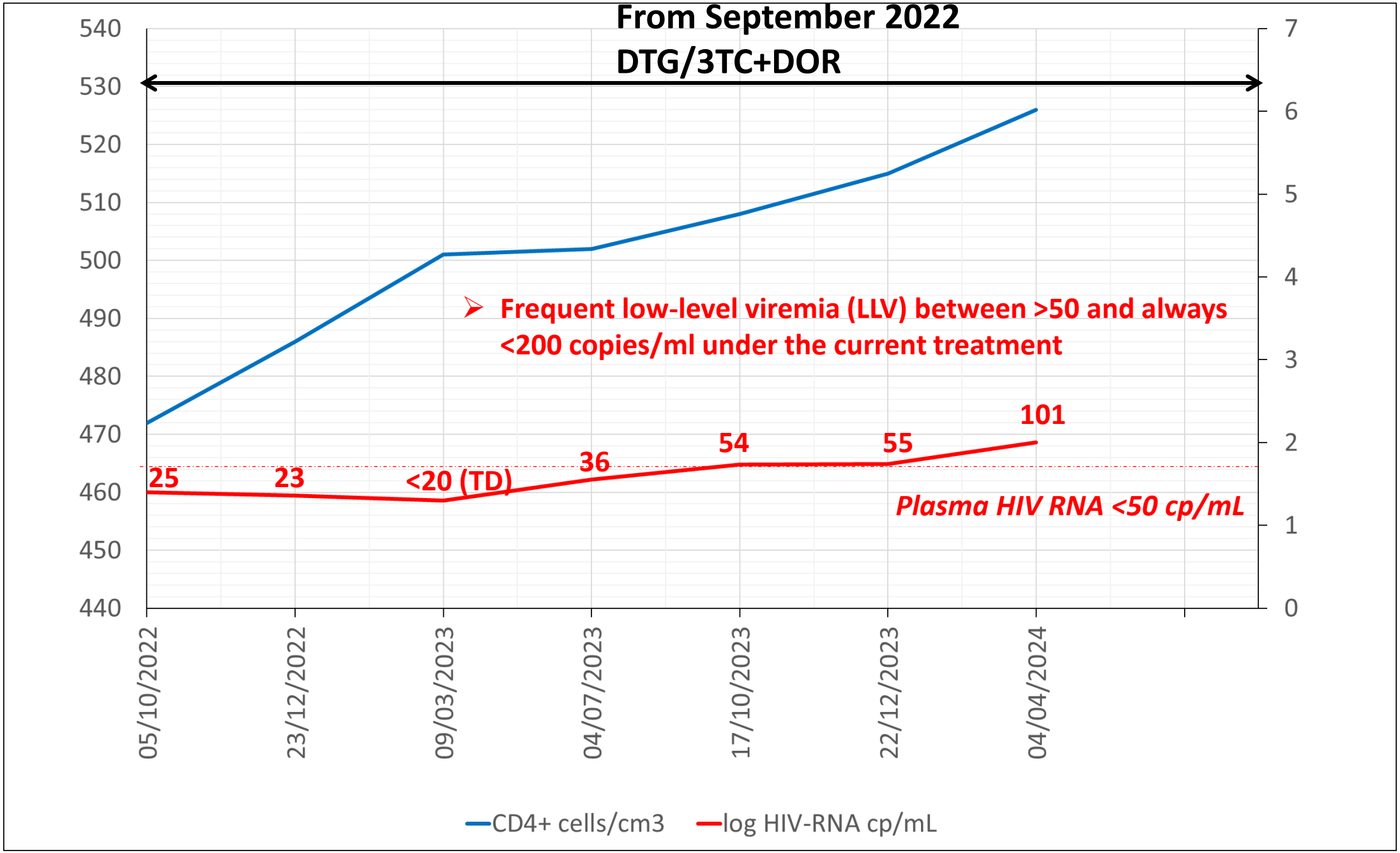
Italian man

- **Non-ART treatments:** Atorvastatin, Omega-3, Vitreoxygen, Dutasteride, Tamsulosin, Cholecalciferol, Zofenopril, Barnidipin
- **cART history:**

1. EFV/TDF/FTC (undefined period from 2000?, autonomously suspended)
2. RAL+TDF/FTC (October 2015 to November 2015 suspended for worsening of kidney functionality)
3. RAL+3TC+DRV/R 800/100mg (November 2015 to November 2016)
4. DTG/3TC+ DRV/R 800/100mg (November 2016 to September 2022)
5. DTG/3TC+DOR (from September 2022)

- **Good adherence**


Oct-Nov 2015
RAL+TDF/FTC
Nov 2015-Nov 2016
RAL+3TC+DRV/R 800/100mg
Nov 2016-Sept 2022
DTG/3TC+DRV/R 800/100mg
DATE: 23/09/2015
GRT ON PLASMA HIV RNA (Sanger)
PR: M36I, L63P, I93L
RT: V179D/V
IN: NONE
DATE: 12/07/2016
GRT ON PLASMA HIV RNA (Sanger)
PR: M36I, L63P, I93L
RT: NONE
IN: NONE
DATE: 08/02/2022
GRT ON HIV DNA (Sanger)
PR: M36I, L63P, I93L
RT: NONE
IN: NONE
Plasma HIV RNA <50 cp/mL
<20 (TND)
<20 (TND)
500
480
460
440
420
400
380
360
340
320
300
280
260
240
220
200
180
160
140
120
100
80
60
40
20
7
6
5
4
3
2
1
0
23/09/2015
27/10/2015
18/11/2015
16/12/2015
12/01/2016
09/02/2016
08/03/2016
11/05/2016
22/06/2016
22/06/2016
12/07/2016
09/08/2016
07/09/2016
16/11/2016
14/12/2016
25/01/2017
06/04/2017
07/08/2017
29/09/2017
13/12/2017
09/04/2018
06/06/2018
03/10/2018
13/02/2019
17/06/2019
25/09/2019
08/01/2020
08/07/2020
11/11/2020
26/02/2021
25/05/2021
23/08/2021
21/12/2021
08/02/2022
23/03/2022
30/06/2022
CD4+ cells/cm3
log HIV-RNA cp/mL


From September 2022
DTG/3TC+DOR
Frequent low-level viremia (LLV) between >50 and always <200 copies/ml under the current treatment
25
23
<20 (TD)
36
54
55
101
Plasma HIV RNA <50 cp/mL
540
530
520
510
500
490
480
470
460
450
440
7
6
5
4
3
2
1
0
05/10/2022
23/12/2022
09/03/2023
04/07/2023
17/10/2023
22/12/2023
04/04/2024
CD4+ cells/cm3
log HIV-RNA cp/mL

No universal agreement but definition is generally set by detectable viremia under failure threshold!

Definitions of plasma HIV-1 RNA detection during ART according to different guidelines

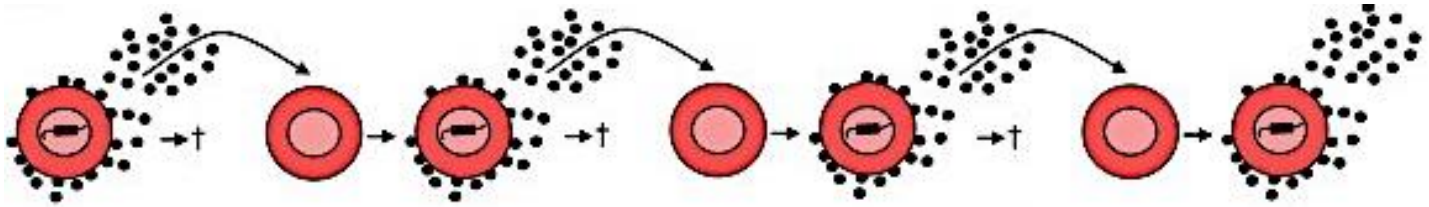
	EACS (1)	JAMA/IAS (2,3)	HIVinfo.gov/DHHS (4)	WHO (5)
Residual Viremia	Not defined	HIV RNA levels >20 and <50 copies/mL (6)	Not defined	Not defined
Low-level Viremia	**HIV RNA levels > 50 and <200 copies/mL**	**HIV RNA levels 50–200 copies/mL (2,3)**	**Confirmed detectable HIV RNA levels <200 copies/mL**	**HIV RNA levels 50–1000 copies/mL**
Viral blip	Not defined	An outlier increase in HIV RNA levels to <1000 copies/mL that returns to undetectable levels (2)	After VS, an isolated detectable HIV-RNA level that is followed by a return to VS	An isolated HIV-RNA measurement of 50–1000 copies/mL with a return to suppressed levels
Viral Rebound	Confirmed HIV RNA levels > 50 copies/mL in someone with previously undetectable viremia	Not defined	After VS, confirmed HIV RNA levels ≥200 copies/mL	Not defined

ART= Antiretroviral Therapy; VS= Viral Suppression.

1. Ambrosioni J, et al. Major revision version 12.0 of the European AIDS Clinical Society guidelines 2023. HIV Medicine. 2023 Nov;24(11):1126–36.
2. Saag MS, et al. Antiretroviral Drugs for Treatment and Prevention of HIV Infection in Adults: 2020 Recommendations of the International Antiviral Society–USA Panel. JAMA. 2020 Oct 27;324(16):1651.
3. Gandhi RT, et al. Antiretroviral Drugs for Treatment and Prevention of HIV Infection in Adults: 2022 Recommendations of the International Antiviral Society–USA Panel. JAMA. 2023 Jan 3;329(1):63.
4. Panel on Antiretroviral Guidelines for Adults N. Guidelines for the Use of Antiretroviral Agents in Adults and Adolescents with HIV How to Cite the Adult and Adolescent Antiretroviral Guidelines 2024. Available from: https://clinicalinfo.hiv.gov/.
5. WHO. Consolidated guidelines on HIV prevention, testing, treatment, service delivery and monitoring: recommendations for a public health approach. 2021. [Available from: https://www.who.int/publications/i/item/9789240031593

Two models to explain low-level viremia

Virus release followed by ongoing virus replication



Ongoing virus replication in sanctuary cellular or body compartments

Virus release with ART halting virus replication

Virus reactivation in latently infected cells in response to stochastic antigenic stimulation, with presence of cART ensuring that new cells cannot be productively infected

In case of LLV, it is important to collect enough information to help differentiate between the two scenarios

- **Which are the causes that determine the persistence of detectable viremia under ART?**

Machanism underlying detectable viremia on ART	Causes
Viral replication (ongoing new infection events)	Suboptimal adherence
	Reduced ART concentration due to drug-to-drug interactions or limited absorbsion
	Drug resistance

Wu F. and Simonetti F., Current HIV/AIDS Reports 2023

Machanism underlying detectable viremia on ART	Causes
Viral replication (ongoing new infection events)	Suboptimal adherence
	Reduced ART concentration due to drug-to-drug interactions or limited absorbsion
	Drug resistance
Viral expression from infected cells (virus production)	Latency reversal resulting in continuous virus production
	Clonal expansion of cells carrying intact or defective proviruses

"Physiological LLV"

Wu F. and Simonetti F., Current HIV/AIDS Reports 2023

- **Which are the consequences of this detectable viremia?**

Machanism underlying detectable viremia on ART	Causes	Consequences
Viral replication (ongoing new infection events)	Suboptimal adherence Reduced ART concentration due to drug-to-drug interactions or limited absorbsion Drug resistance	Viral evolution Drug resistance Increase in viremia and virological failure $CD4^+$ T cells loss Risk of transmission

Wu F. and Simonetti F., Current HIV/AIDS Reports 2023

Machanism underlying detectable viremia on ART	Causes	Consequences
Viral replication (ongoing new infection events)	Suboptimal adherence	Viral evolution
	Reduced ART concentration due to drug-to-drug interactions or limited absorbsion	Drug resistance
		Increase in viremia and virological failure
		$CD4^+$ T cells loss
	Drug resistance	Risk of transmission
Viral expression from infected cells (virus production)	Latency reversal resulting in continuous virus production	No viral evolution
		No drug resistance to current regimen
		Intermittent or stable viremia for months/years
	Clonal expansion of cells carrying intact or defective proviruses	Stable $CD4^+$ T cells
		Potential higher inflammation

Wu F. and Simonetti F., Current HIV/AIDS Reports 2023

Under these conditions of LLV, which of the following intervensions do you intend to use to manage the situation?

a) Change therapy immediately

b) Evaluating resistance

c) Evaluating adherence

d) Evaluation of HIV DNA levels

A considerable resistance is also observed at LLV

HIV/AIDS MAJOR ARTICLE

Reliability and Clinical Relevance of the HIV-1 Drug Resistance Test in Patients With Low Viremia Levels

Maria Mercedes Santoro,[1] Lavinia Fabeni,[2] Daniele Armenia,[1] Claudia Alteri,[1] Domenico Di Pinto,[1] Federica Forbici,[2] Ada Bertoli,[1,3] Domenico Di Carlo,[1] Caterina Gori,[2] Stefania Carta,[2] Valentina Fedele,[2] Roberta D'Arrigo,[2] Giulia Berno,[2] Adriana Ammassari,[2] Carmela Pinnetti,[2] Emanuele Nicastri,[2] Alessandra Latini,[4] Chiara Tommasi,[2] Evangelo Boumis,[2] Nicola Petrosillo,[2] Gianpiero D'Offizi,[2] Massimo Andreoni,[1,3] Francesca Ceccherini-Silberstein,[1] Andrea Antinori,[2] and Carlo Federico Perno[1,2,3]

In PWH failing cART with LLV, Sanger HIV-1 genotyping provides reliable and reproducible results that are informative about emerging drug resistance.

HIV-1 drug resistance stratified by plasma viremia ranges

	All Samples				
	Resistance to Any Class				
Viremia Range, Copies/mL	No.	%	*P* Value[a]	PRMs, No.	*P* Value[b]
Overall ranges	3895	74.0		3 (0–7)	
50–200	396	52.8	<.001	1 (0–3)	<.001
201–500	287	70.0		2 (0–4)	
501–1000	242	74.0		3 (0–6)	
1001–10 000	1102	86.1		4 (2–7)	
10 001–100 000	1212	76.7		4 (1–8)	
>100 000	656	63.0		2 (0–8)	

Santoro et al. CID 2014

Genotypic Resistance Testing in Clinical Practice: which to use

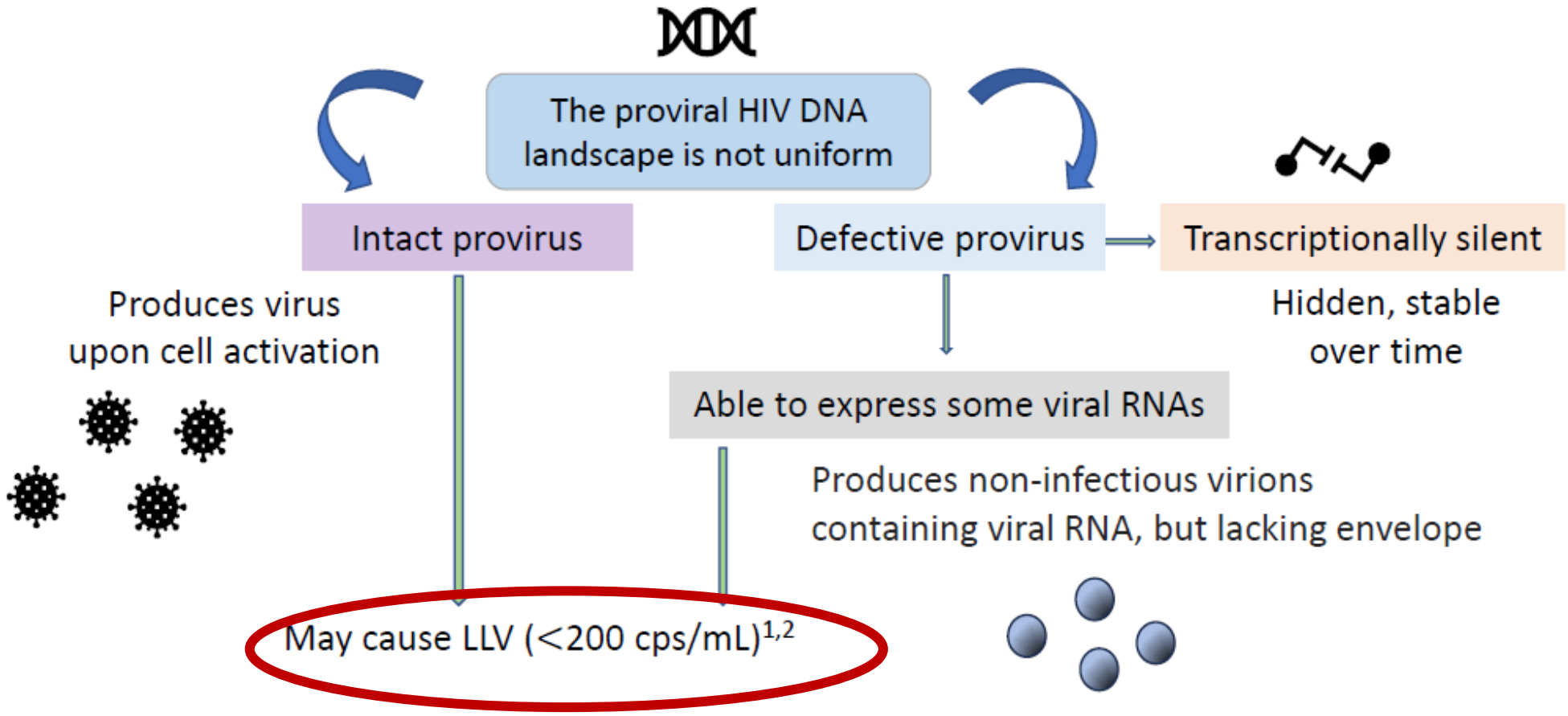
- Genotypic resistance testing (GRT) through Sanger sequencing of plasma HIV RNA has been long and effectively supporting ART.

- In individuals **under virological control or with low-level viremia**, GRT performed on plasma virus cannot be consistently performed, while **GRT on peripheral blood mononuclear cells (PBMCs) is technically feasible** and can represent **a valuable tool for the diagnosis of resistance**.

  - ✓ Sanger or NGS?

  - ✓ Advantages and disadvantages, always keeping in mind the risk of overestimation (*low abundance mutations, APOBEC editing, non-replicating HIV-DNA*) or underestimation (*technical/temporal issues, low level HIV-DNA, missing previously existing drug-resistance mutations*).

The HIV DNA reservoir during suppressive ART is associated with LLV



Adapted from Geretti AM. Infect Dis Ther 2022; [1]Cole et al. Cell Rep 2022; [2]White et al. J Clin Invest 2023

HIV-DNA= 3275 copies/10^6 CD4 DATE: 18-01-2024
CD4=523 cells/mm^3 (27%)

GRT ON HIV- DNA NGS 5%

KNOWN MUTATIONS*:
PROTEASE: M36I (100%), L63P (99%), I93L (100%)
REVERSE TRANSCRIPTASE: NONE
INTEGRASE: NONE

APOBEC MUTATIONS*:
PR: NONE
RT: G18G/R (G94%, R 5%), G51G/R (G93%, R 7%), W88W/* (* 6%, W94%), G93G/R (G93%, R6%), W153W/* (* 6%, W93%)
IN: NONE

Mutations deriving from APOBEC activity could interfere with the full replicative capacity of the virus, making it more difficult, and indirectly supporting the effectiveness of the therapy

RESULT:
PR: Presence of secondary mutations
RT: Absence of mutations conferring resistance to RTIs
IN: Absence of mutations conferring resistance to Integrase inhibitors

TROPISM: Virus with X4 tropism (FPR: 1.7%)

**http://hivdb.stanford.edu/*

POTENTIALLY EFFECTIVE DRUGS:
PR: All
RT: All
IN: All
CCR5: None

NOTE: Test performed with ultrasensitive sequencing (NGS) method for the recognition of minority species ≥ 5%. V3 performed using Sanger method.

VIROLOGICAL INTERPRETATION: In the present genotypic picture, obtained on lymphomonocyte DNA, the following is observed:
-In Protease, absence of resistance mutations;
-In Reverse Transcriptase, compared to the latest DNA genotype of 2022, the disappearance, even as minority variant, of the V179D/V mutation (present on plasma in the 2015 genotype) is confirmed, capable of modulating the full effectiveness of NNRTIs. Additionally, we note, in a minority form, the presence of **5 APOBEC related mutations**, two of which are stop codons.
-In Integrase absence of resistance mutations.
In conclusion, the presence of mutations deriving from the function of APOBEC are indicative of a defective virus (Armenia D. et al., J Clinical Virology 2023) interfering with the full replicative capacity of the virus.
Therefore, the residual viremia that is still detectable seems more to be attributed to the release of viral particles from infected cells rather than to the presence of true replicative cycles.

NO RESISTANCE MUTATIONS
NO HISTORY OF VIROLOGICAL FAILURES SINCE 2015

In February 2024:

Abdominal ultrasound:
mild hepatic steatosis
polycystic kidney disease
prostatic hypertrophy
Echo of the supra-aortic trunks:
plaque causing mild carotid stenosis < 20%

BMI: 28.69
Bone densitometry:
Femur -1.2 SD; Lumbar: 0.9 SD
FRAX Score: 2.7% in 10 years
Heart SCORE2-OP: 24% in 10 years

Laboratory values:
eGFR:42.8 ml/min
Creatinine 1.53 mg/dl
Cholesterol (total) 168 mg/dl
Cholesterol (LDL) 113 mg/dl
Cholesterol (HDL) 37 mg/dl
Triglycerides 117 mg/dl

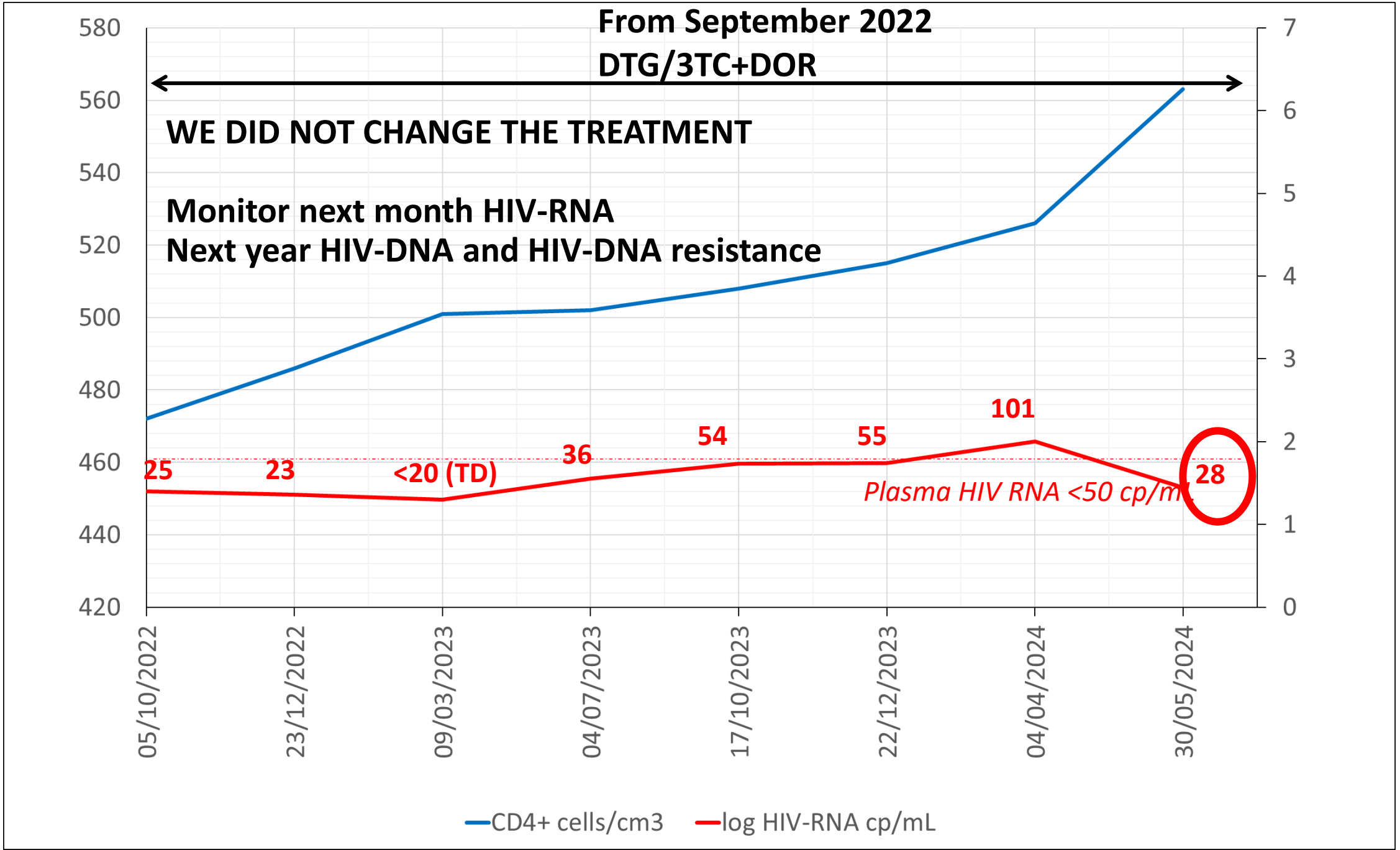
WHAT TO DO?

a) Change therapy to BIC/TAF/FTC

b) Change therapy to DOR/TDF/3TC

c) Change therapy to DTG/3TC

d) No change therapy


From September 2022
DTG/3TC+DOR
WE DID NOT CHANGE THE TREATMENT
Monitor next month HIV-RNA
Next year HIV-DNA and HIV-DNA resistance
25
23
<20 (TD)
36
54
55
101
28
Plasma HIV RNA <50 cp/mL
580
560
540
520
500
480
460
440
420
7
6
5
4
3
2
1
0
05/10/2022
23/12/2022
09/03/2023
04/07/2023
17/10/2023
22/12/2023
04/04/2024
30/05/2024
CD4+ cells/cm3
log HIV-RNA cp/mL

Clinical management recommendations for PHW with LLV

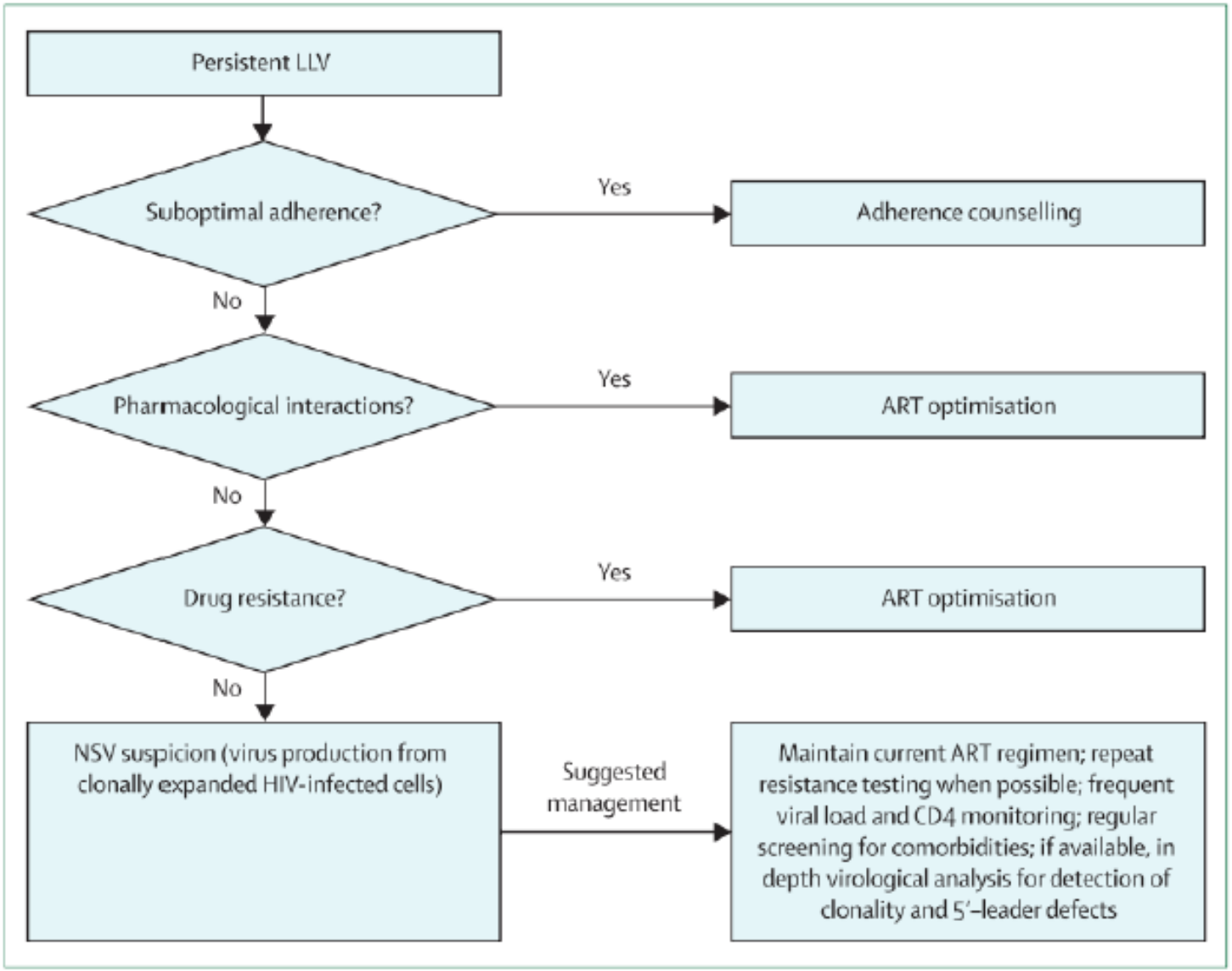


Figure: **Clinical management recommendations for patients with persistent LLV and suspected of NSV**
ART=antiretroviral therapy. LLV=low-level viremia. NSV=non-suppressible viremia.

Lancet HIV 2024; 11: e333–40

Published Online
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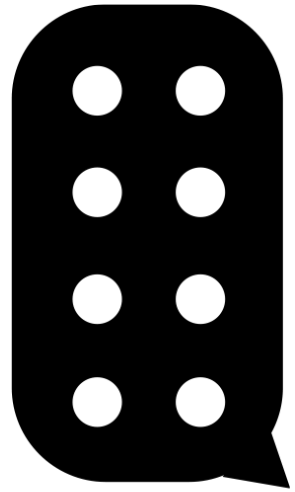
S2352-3018(24)00063-8

Thank you for the attention



HIV CLINICAL TOPICS

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