ZKBS
The Central Committee on Biological Safety

MEACB 2019
9th Meeting of the European Advisory Committees on Biosafety
in the field of contained use and deliberate release of GMOs
6-7 November 2019, Berlin, Germany

Federal Office of Consumer Protection and Food Safety

Details from a special case: laboratory acquired HIV-1 infection

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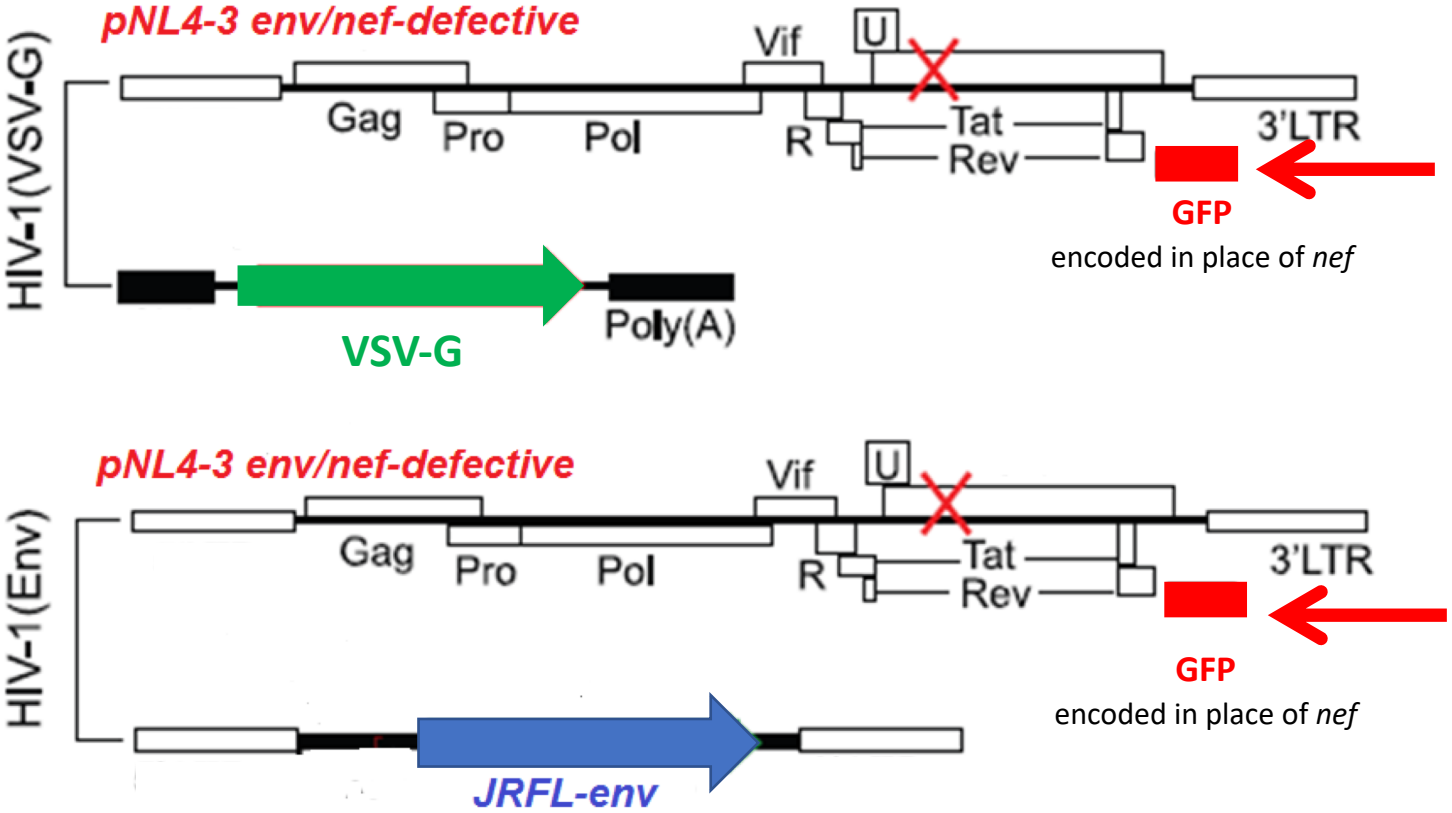
Case report

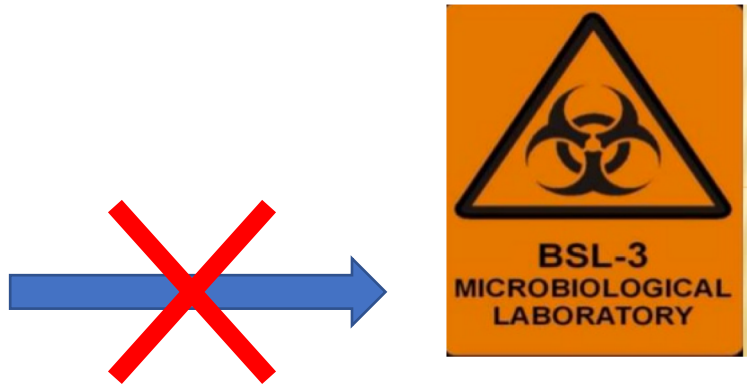
- A blood donor was found to be HIV-1 infected at routine screening
- Previous tests (the latest 7 months before) were negative
- No classical risk factors for HIV acquisition:
  - The unique partner was tested HIV-negative
  - No blood transfusions
  - No piercing or tattoos
  - No injecting drug use
  - No invasive medical procedures

However, during the 6 months preceding HIV diagnosis…

The subject worked in a research laboratory, with the exclusive task of producing pseudoviruses by co-transfecting 293T cells with HIV-1 **NL4-3 env/nef-defective**, green fluorescent protein–labeled vector, and **JRFL Env**- or **VSV-G** encoding plasmid in **BSL-2**



No access to BSL-3 was allowed or reported



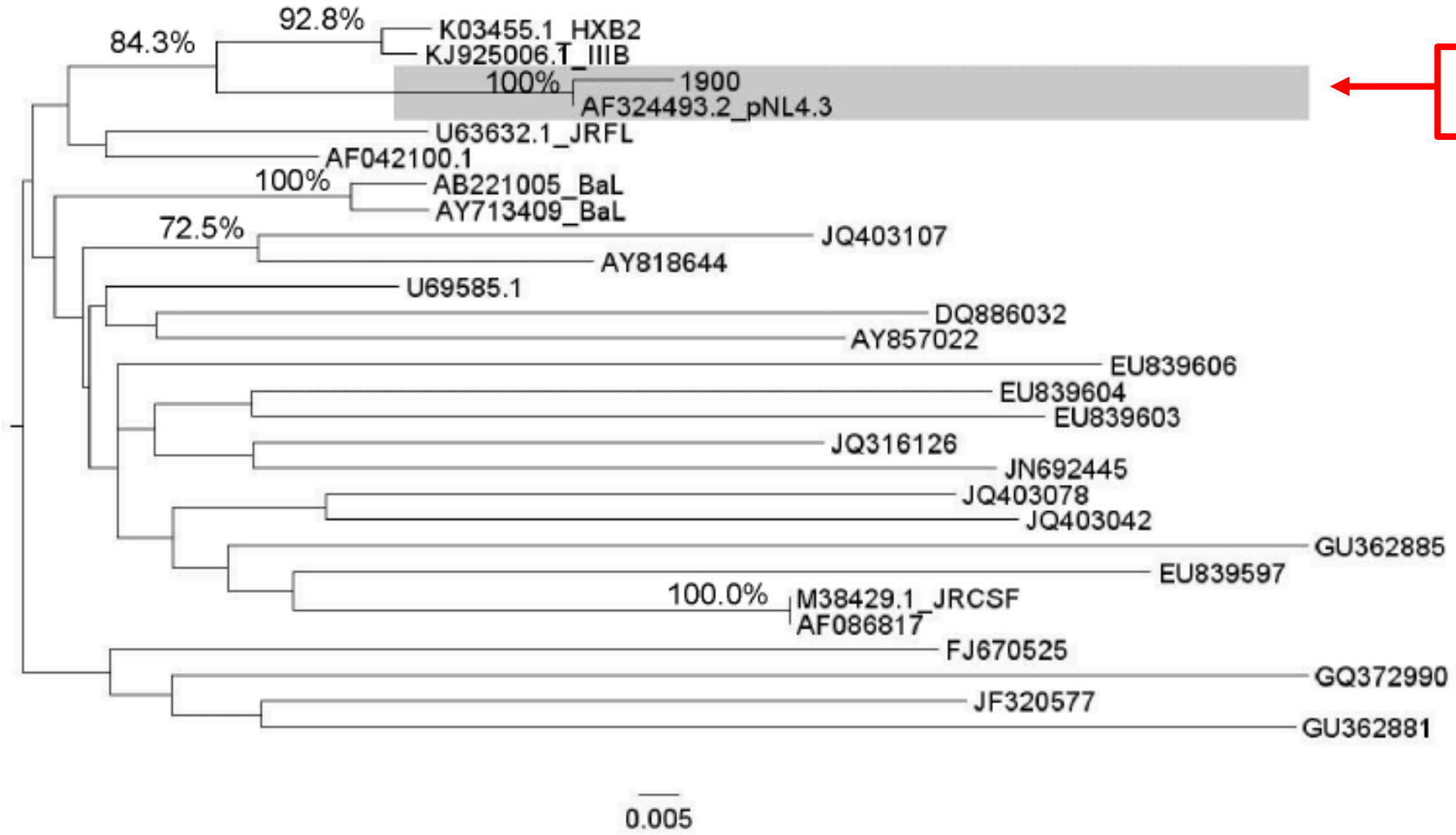
No major or minor laboratory **accidents** (percutaneous injury, medium splash, breaking of gloves, eye or skin contact with cell-derived medium, etc) were recalled

Methods

- Whole genome HIV-1 sequencing
- Phylogenetic analysis
- Viral isolation
- Phenotypic determination
- Neutralizing activity assessment
- Viral evolution analysis

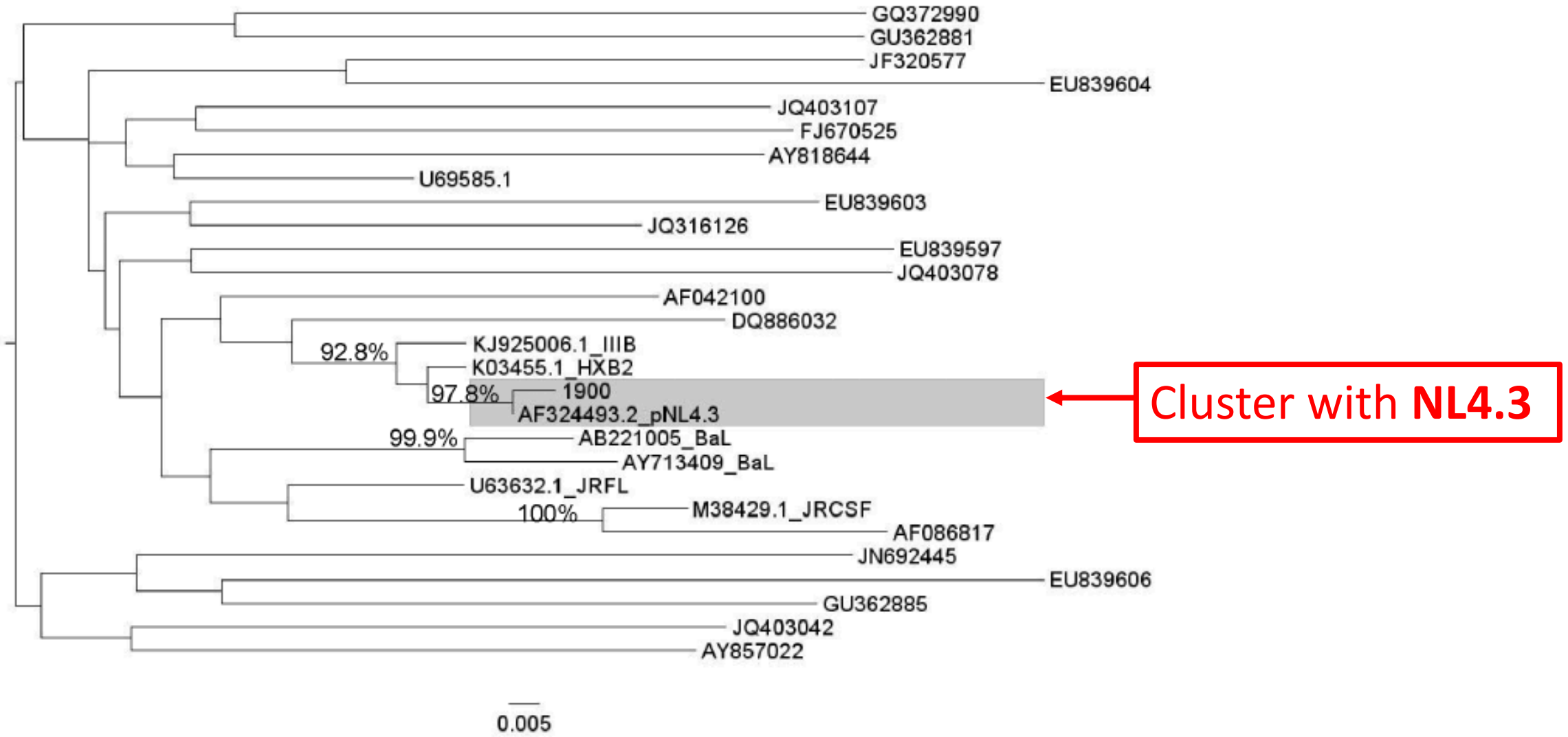
Phylogenetic tree for gag-pol

Phylogenetic tree for Vif, Vpr, Vpu, Tat/Rev

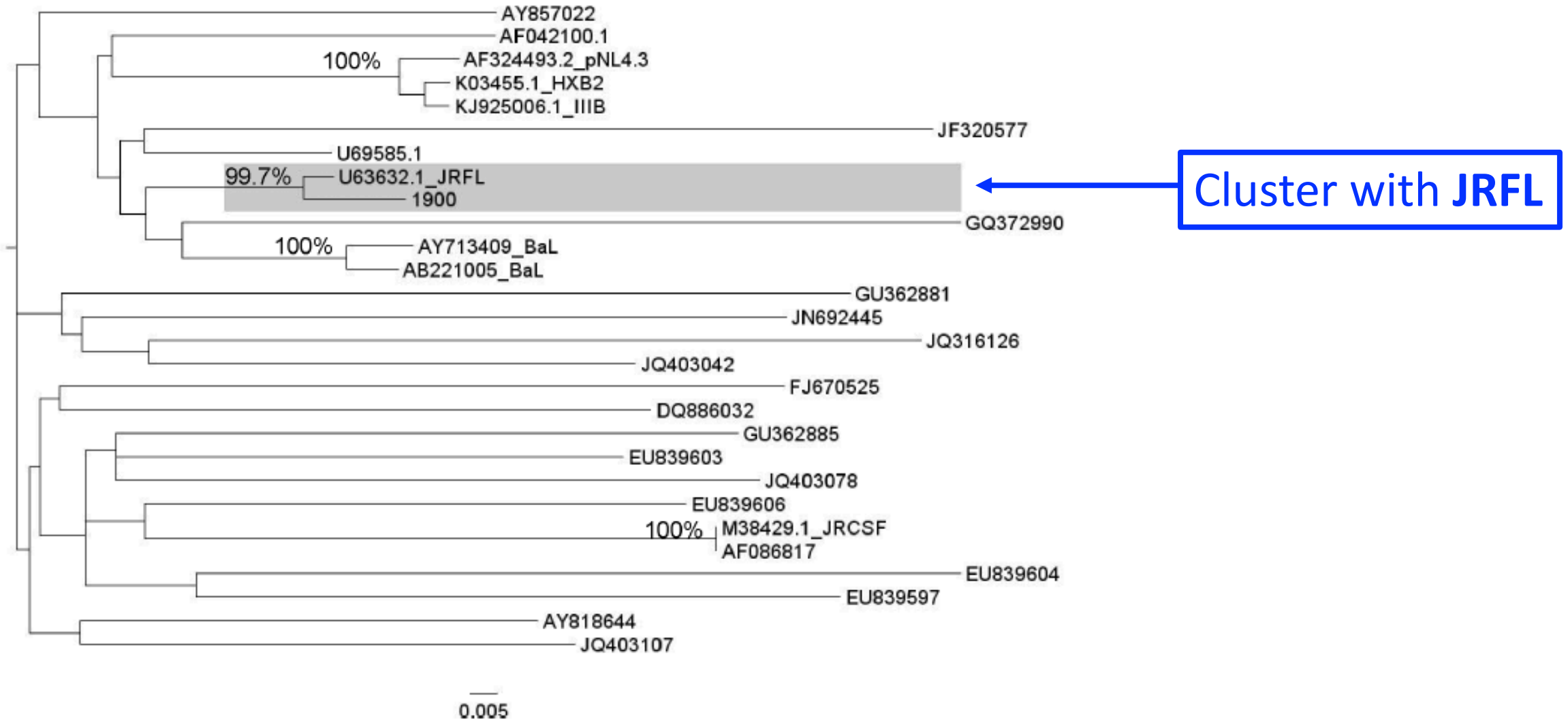


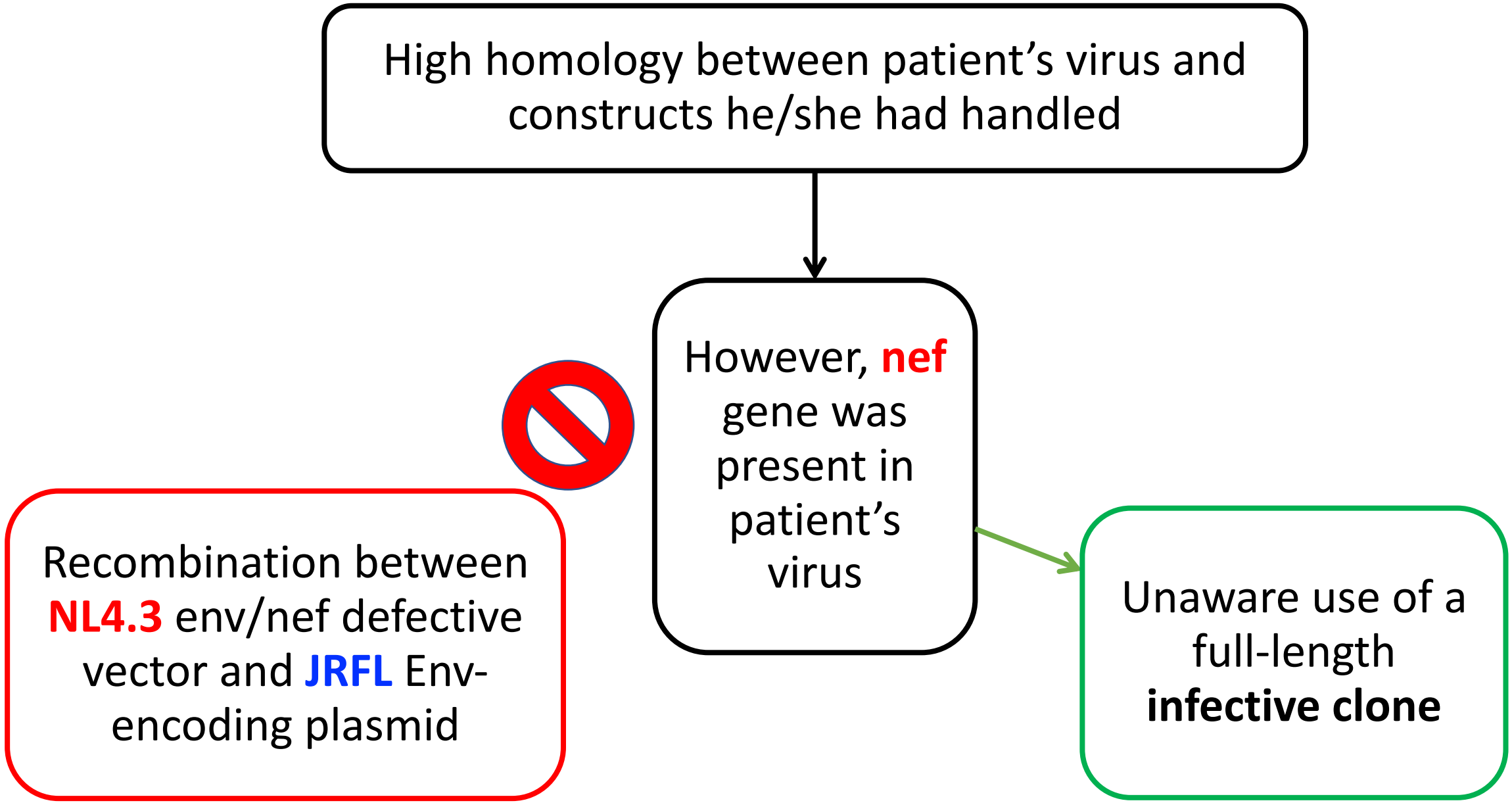
Cluster with **NL4.3**

Phylogenetic tree for nef

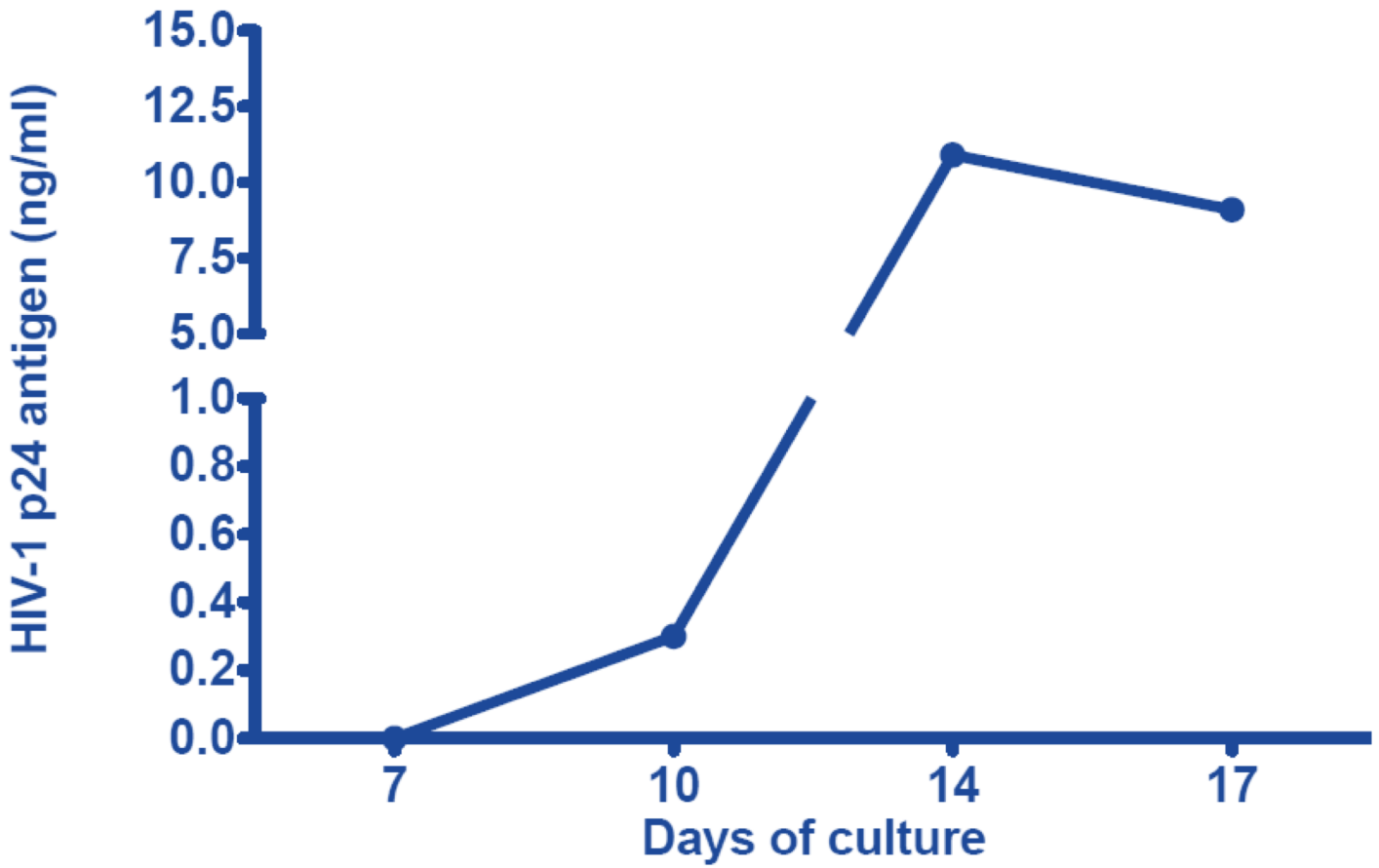


Phylogenetic tree for Env




High homology between patient's virus and constructs he/she had handled
However, nef gene was present in patient's virus
Recombination between NL4.3 env/nef defective vector and JRFL Env-encoding plasmid
Unaware use of a full-length infective clone

Viral isolation



Determination of viral tropism

Phenotyping

	HIV p24-Ag (ng/ml day 7)
U87.CD4.CCR5	34 (++ Syncytia)
U87.CD4.CXCR4	4.9 (no Syncytia)

Virus sensitivity to antibodies (autologous)

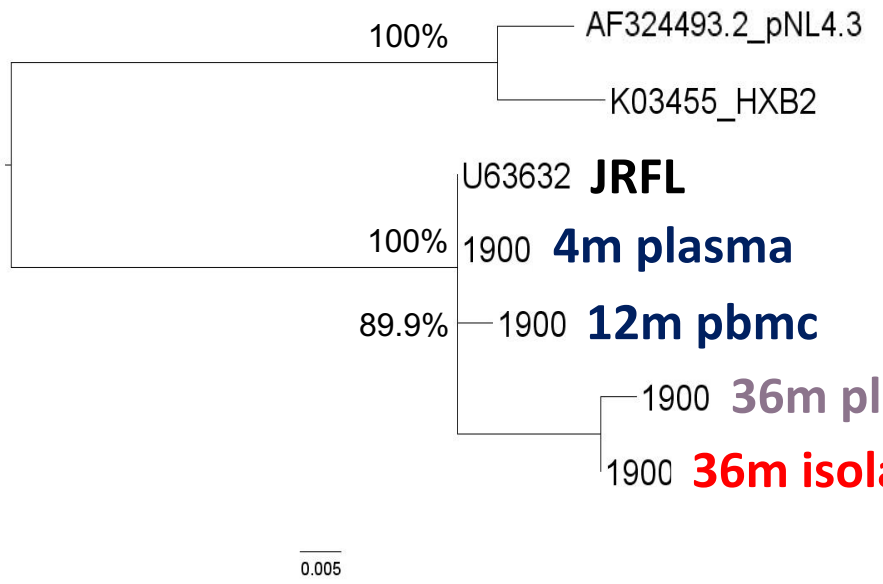
HIV-1 Viruses	**Serum neutralization (IC50) in Tzm-bl assay (ug/ml)**
Primary virus #1900	<20
JRFL (B)	495
Tier 1	
SF162 (B)	916
93MW965 (C)	1,397
GS015.EC12 (C)	304
TH023 (CRF01)	40
Tier 2	
DU151 (C)	<20
CM235 (CRF01)	<20
Control	
VSV-G	<20

Virus sensitivity to monoclonal antibodies

	Neutralization (IC50) with MoAbs in Tzm-bl assay (ug/ml)					
Virus	**PG9**	**PG16**	**b12**	**2G12**	**4E10**	**2F5**
Primary virus #1900	**0.0049**	**0.0059**	**3.37**	**>5**	**>5**	**>5**
JRFL (B)	**>5**	**>5**	**0.014**	**1.21**	**3.77**	**0.78**

Viral evolution

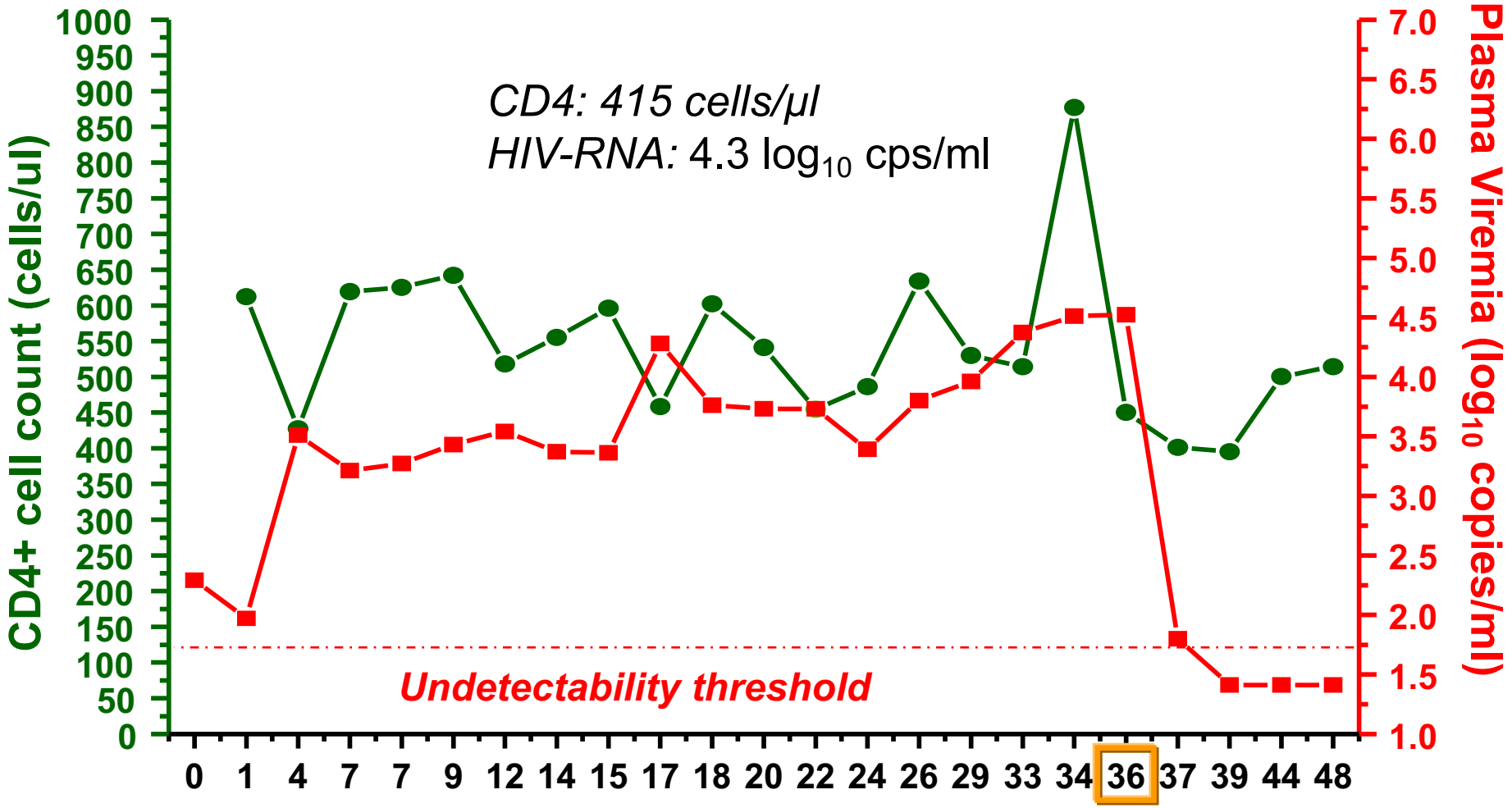
A.



B.

dN/dS	*Env* N-glycosylation sites	FPR (%)
-	28^{TOT}	24.7
0	*na*	24.7
1	+1 (C2) 29^{TOT}	27.1
2.2	+1 (C2) 29^{TOT}	17.0
2	*na*	8.1

Clinical follow-up



Finally: what's really happened?

We do not know yet. We can only make some hypotheses:

1. Labelling error

2. Unaware handling of infective clone in co-transfection experiments with VSV-G plasmid

3. Unaware contact with mucous membrane of a high-titer replicating HIV-1 culture

Lesson learned

- Trust the patient!
- Implement procedures to avoid contamination between BSL-2 and BSL-3
- Strict procedures of clone labelling
- …

Acknowledgements

University of Rome "Tor Vergata"

C. Alteri
C.F. Perno
F. Ceccherini-Silberstein
A. Bertoli
F. Continenza
M. Bellocchi
E. Balestra
M. Bruni
D. Armenia
L. Carioti
M. Surdo
P. Saccomandi

San Gerardo Hospital

A. Gori
A. Bandera
I. Beretta
S. Malandrin

San Raffaele Scientific Institute

G. Scarlatti
M. Tolazzi

& the patient

For futher detais see publication: *Soria A, Alteri C, et al. Occupational HIV infection in a research laboratory with uknown mode of transmission: a case report. Clin Infect Dis 2017;64(6):810-3*