

Dynamic of HIV-1 RNA and DNA in plasma and cervico-vaginal secretions after Highly Active Antiretroviral Therapy initiation: ANRS EP24 GYNODYN Study

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BACKGROUND

HIV shedding in cervico-vaginal secretions is a major determinant of sexual HIV transmission

A better understanding of the viral dynamics in this compartment in relation with antiretroviral treatment may improve the prevention of HIV transmission.

OBJECTIVES

We studied the dynamic of HIV-RNA and DNA levels in plasma and cervico-vaginal secretions over a period of 18-months after HAART-initiation among HAART-naïve HIV-1-infected women in France.

METHODS

Design: prospective multicentric open cohort of 23 antiretroviral (ARV)-naïve women with HIV-1 infection initiating zidovudine/lamivudine combined with lopinavir (n=17) or indinavir (n=6) boosted with low-dose ritonavir.

Laboratory method

Paired samples of plasma and cervico-vaginal secretions (obtained by cervico-vaginal lavage) were collected at baseline, then at 1, 6, 12 and 18 months to measure viral markers and antiretroviral concentrations.

Viral HIV-RNA and proviral HIV-DNA were measured using real-time PCR assay. The threshold of detection was 50 copies/ml ($1.7 \log_{10}$) for viral RNA and 10 copies/ 10^6 cells ($1.0 \log_{10}$) for proviral DNA.

Antiretroviral concentrations were measured by high performance liquid chromatography at different points in time after antiretroviral intake.

Viral HIV-RNA and antiretroviral concentration from cervico-vaginal secretions were adjusted using a gold standard solution of Lithium.

Statistical methods

Mean HIV-RNA and proviral DNA in plasma and cervico-vaginal secretions after \log_{10} transformation were described in the two compartments.

We compared mean HIV-RNA in plasma and dilution factor adjusted HIV-RNA in cervico-vaginal secretions using a Wilcoxon non parametric test, all HIV-RNA values lower than the adjusted cervico-vaginal secretions threshold being replaced by $2.7 \log_{10}$ cells.

RESULTS

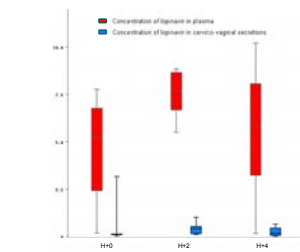
Table 1: Baseline characteristics (N=23)
ANRS EP24 Gynodyn, 2003-2006

Characteristics	n	%
Median age in years (IQR)	33	(30-35)
Transmission group, n (%)		
sexual	20	(87)
unknown	3	(13)
Body Mass Index in kg/m^2 , n (%)		
<19	1	(5)
[19-25[10	(50)
[25-30[7	(35)
≥ 30	2	(10)
Clinical stage, n (%)		
A	18	(78)
B	2	(9)
C	3	(13)
Using effective method of contraception	20	(87)
ARV treatment		
lopinavir/ritonavir	17	(74)
indinavir/ritonavir	6	(26)
Median CD4+ cells/ μl (IQR)	199	(128-310)
Median CD8+ cells/ μl (IQR)	790	(486-1055)
Mean HIV RNA in plasma in \log_{10} copies/ml (SD)	4.74	(0.96)
Mean HIV DNA in cervico-vaginal secretions in \log_{10} copies/ml (SD)	3.89	(1.01)
Mean HIV DNA in plasma in \log_{10} / 10^6 cells (SD)	3.52	(0.50)
Mean HIV DNA in cervico-vaginal secretions in \log_{10} / 10^6 cells (SD)	2.02	(0.88)

Abbreviations: IQR Interquartile range
SD Standard deviation

PHARMACOLOGICAL RESULTS

Figure 1: Concentration of lopinavir at Month 1 (N=15),
ANRS EP24 Gynodyn, 2003-2006



Boxplots (median, IQR) of concentration of lopinavir in plasma and in cervico-vaginal secretions (dilution factor adjusted) at medication time (H+0), two hours after medication (H+2) and four hours after medication (H+4)

RNA HIV RESULTS

Table 2: Evolution of HIV RNA in plasma and in cervico-vaginal secretions
ANRS EP24 Gynodyn, 2003-2006

	Day0	Month1	Month6	Month12	Month18
Number of available samples (%)					
plasma	22 (96)	18 (78)	14 (61)	9 (39)	8 (35)
cervico-vaginal secretions	22 (96)	18 (78)	14 (61)	10 (43)	9 (39)
Number of undetectable values (%)					
plasma (< 1.7 \log_{10} copies/ml)	1 (5)	3 (17)	10 (71)	6 (67)	6 (75)
plasma (= 1.7 and < 2.7 \log_{10} copies/ml) (censored)	0	4 (22)	4 (29)	0	0
cervico-vaginal secretions (< 2.7 \log_{10} copies/ml)	3 (14)	14 (78)	13 (93)	9 (100)	8 (100)
Mean HIV-1 RNA in \log_{10} copies/ml (SD)					
plasma*	4.79 (0.82)	3.15 (0.75)	2.70 (0.00)	3.04 (0.52)	3.30 (1.11)
cervico-vaginal secretions*	3.95 (0.92)	2.90 (0.64)	2.81 (0.37)	2.81 (0.19)	2.94 (0.34)
p-value ^c	0.0008	0.0077	0.5000	0.8125	1.0000

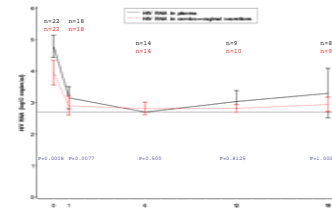
Abbreviations: SD Standard deviation

a) all HIV-RNA values lower than the adjusted cervico-vaginal secretions threshold were replaced by $2.7 \log_{10}$

b) dilution factor adjusted

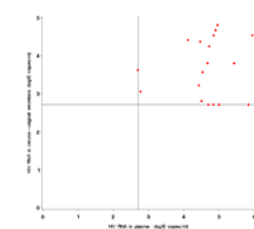
c) adjusted Student T or Wilcoxon test of plasma versus cervico-vaginal HIV-1 RNA

Figure 2: Evolution of mean HIV RNA in plasma and in cervico-vaginal secretions,
ANRS EP24 Gynodyn, 2003-2006



Vertical bars represent 95% confidence intervals, the horizontal line the detection limit ($2.7 \log_{10}$).
P values are results of paired T tests or paired Wilcoxon signed rank sum tests.

Figure 3: Correlation between HIV RNA in cervico-vaginal secretions and in plasma
at baseline, ANRS EP24 Gynodyn, 2003-2006



Vertical and horizontal lines represent detection limits ($2.7 \log_{10}$).
Pearson's correlation coefficient between HIV-1 RNA in cervico-vaginal secretions and in plasma $p=0.3409$ $p=0.1205$.

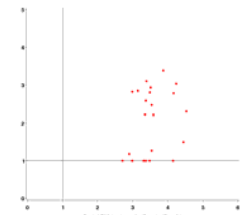
DNA HIV RESULTS

Table 3: Evolution of HIV DNA in plasma and in cervico-vaginal secretions, ANRS EP24 Gynodyn, 2003-2006

	Day0	Month1	Month6	Month12	Month18
Number of available samples (%)					
plasma	23 (100)	17 (74)	11 (48)	10 (43)	6 (26)
cervico-vaginal secretions	23 (100)	18 (78)	15 (65)	10 (43)	9 (39)
Number of undetectable values (%)					
plasma (< 1 \log_{10} cp/ 10^6 cells)	0	0	0	0	0
cervico-vaginal secretions (< 1 \log_{10} cp/ 10^6 cells)	7 (30)	11 (61)	8 (53)	7 (70)	5 (56)
Mean HIV-1 DNA in \log_{10} copies/10^6 cells (SD)					
plasma	3.52 (0.50)	3.29 (0.63)	2.77 (0.59)	2.83 (0.47)	2.80 (0.30)
cervico-vaginal secretions	2.02 (0.88)	1.47 (0.67)	1.58 (0.83)	1.33 (0.55)	1.30 (0.52)

Abbreviations: SD Standard deviation

Figure 4: Correlation between HIV DNA in cervico-vaginal secretions and in plasma at baseline, ANRS EP24 Gynodyn, 2003-2006



Vertical and horizontal lines represent detection limits ($1 \log_{10}$).
Pearson's correlation coefficient between HIV DNA in cervico-vaginal secretions and in plasma $p=0.2778$ $p=0.1994$.

DISCUSSION - CONCLUSION

• Baseline viral markers were significantly lower in cervico-vaginal secretions than in plasma and were not correlated with each other

• HIV-RNA decreased significantly in both body compartments after HAART initiation reaching durable undetectable values from one-month up to 18 months.

• By contrast, HIV-DNA levels remained detectable in all plasma samples and most cervico-vaginal secretions samples over time.

• Concentration of lopinavir in cervico-vaginal secretions was lower than in plasma.