ARCHIVED HIV-1 DNA MUTATIONS TO RILPIVIRINE IN SUBJECTS PREEXPOSED TO EFAVIRENZ OR NEVIRAPINE

Poster 591

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Background

- Failure to the first generation of NNRTI (efavirenz and nevirapine) is associated to a rapid selection of resistance-associated mutations (RAMs) in plasma
- In patients pre-exposed to efavirenz/nevirapine but now controlled for many years with antiretroviral treatment (ART), the switch to second generation of NNRTI (as rilpivirine or etravirine) may be considered
- The use of past plasma resistance genotypes is recommended to determine the rilpivirine susceptibility but these are not always available
- HIV-1 DNA is archived as integrated proviruses in infected cells (actively replicating or latently infected) and can act as a record of drug resistance mutations
  - Peripheral blood mononuclear cells (PBMCs) are a source of archived HIV-1 DNA and can be used for genotypic analyses
  - Transmitted or previously emergent drug resistance mutations may be detectable in the proviral HIV-1 DNA of circulating PBMCs from blood that has <50 copies/mL of HIV-1 RNA
- The virological response to ART may also be impacted using regimen containing drugs for whom specific archived resistance in HIV DNA have been detected

Study objective

- To determine the prevalence of RAMs to rilpivirine, etravirine and to the combination of tenofovir/emtricitabine/rilpivirine on HIV1 DNA on patients previously exposed to efavirenz or/and nevirapine

Methods

- All 169 patients enrolled in the ANRS 138-EASIER trial had already received three antiretroviral drug classes (NRTI, NNRTI and PI) and had plasma HIV-1 RNA < 400 cp/ml at randomization
- From whole blood, reverse transcriptase gene (RT) sequencing was performed on HIV-1 DNA at the time of inclusion
- We analysed the pattern and the prevalence of RAMs to rilpivirine, etravirine, tenofovir and emtricitabine
- Between groups comparisons were carried out using the Wilcoxon test.

Genotyping methods

- HIV-1 DNA resistance genotyping was performed in a central laboratory prior to randomization
- Viral DNA was extracted from 200 µL of frozen stored whole blood using an automatic nucleic acid extractor (MagnaPure; Roche, Meylan, France)
- Reverse transcriptase–polymerase chain reaction (RT-PCR) and then nested PCR were used to amplify the reverse transcriptase (RT) and protease (PR) genes according to ANRS consensus methods (www.hivfrenchresistance.org)
- Population sequencing was performed on purified amplicons with the Taq Dye Deoxy Terminator cycle sequencing kits (Applied Biosytems, Courtaboeuf, France) and resolved on an ABI 3700 automated DNA sequencer (Applied Biosytems, Courtaboeuf, France)
- Sequences were aligned with the HIV-1 subtype B HXB2 reference strain(SmartGene, Lausanne, Switzerland).

Resistance mutations

- Resistance to antiretroviral drugs was evaluated according to the September 2013 ANRS algorithm (www.hivfrenchresistance.org) and the March 2013 International AIDS Society list (www.iasusa.org):
  - **To rilpivirine**, as having at least one mutation among K101E/P, E138A/G/K/Q/R/S, V179L, Y181C/I/V, Y188L, H221Y and M230I/L/V, or as having L100I + K103N/S, L100I + K103R + V179D
  - **To etravirine**, as having at least 4 among: V90I, A98G, L100I, K101E/H/I/P/R, V106I, V179D/F/I/L/M/T, Y181C/I, G190A/S, M230L, or as having at least one mutation among E138K, Y181V and Y181C+H221Y
  - **To tenofovir**, as having at least 6 mutations among M41L, E44D, D67N, T69D/N/S, L74V/I, L210W and, or as having at least one mutation among K65R/E, insertion at codon 69 and K70E
  - **To emtricitabine**, as having at least one mutation among M184V/I and insertion at codon 69

Results

- RT gene amplification succeeded in 128/169 (76%) patients. No difference in CD4 cell count was observed between patients in whom amplification succeed or failed (Table 1).

Table 1: RT amplification success according to baseline CD4 cell count

Baseline CD4 (/mm³)	*RT DNA amplification*		*Total*	*p*
	Success (n=128)	*Failure (n=41)*	*(n=169)*	
Min-Max	113 to 1254	87 to 872	87 to 1254	
Median [IQR]	395 [270;513]	364 [241;512]	393 [267;512]	0.50

Table 2: Baseline characteristics (n=128)

Age	Min-Max	19.9 to 70.8
(years)	Median [IQR]	48.4 [43.3 ; 56.2]
Sex	Male	108 (84%)
CDC Classification	C	63 (49%)
Baseline CD4	Min-Max	113 to 1254
(/mm³)	Median [IQR]	395 [270 ; 513]
Nadir CD4	Min-Max	0 to 404
(/mm³)	Median [IQR]	56 [14 ; 118]
Baseline ART duration	Min-Max	4.4 to 20.1
(years)	Median [IQR]	13.6 [12 ; 15.2]
ART at baseline		
NRTI	All	122 (95%)
	TDF	73 (57%)
	3TC/FTC	97 (76%)
	ABC	48 (38%)
NNRTI		7 (5%)
PI		127 (99%)
RNA HIV-1 (<50 cp/mL)	no. (%)	107 (84%)
HIV-1 subtype	B	122 (95%)
	Non B	6 (5%)

- Patients were treated for a median of 13.6 years and were controlled for at least 2 years.
- **Rilpivirine RAMs** were detected in **40/128 (31%)** of patients, with highest frequency for the mutations Y181C/I/V (23/40, 18%), K101E/P (9/40, 7%) and E138A/G/K/Q/R/S (8/40, 31%) (Figure 1).
- **Etravirine RAMs** were detected in **5/128 (4%)** of patients, with highest frequency for the mutations profiles Y181C+H221Y (2/5, 2%) and the etravirine RAMs set (2/5, 2%) (Figure 2).
- **Resistance to emtricitabine and tenofovir** were detected in 72/128 (56%) and 12/128 (9%), respectively (Figure 3).
- **Resistance to at least one drug included in the combination of tenofovir/emtricitabine and rilpivirine** was observed for **88/128 (69%)** patients (Table 3).

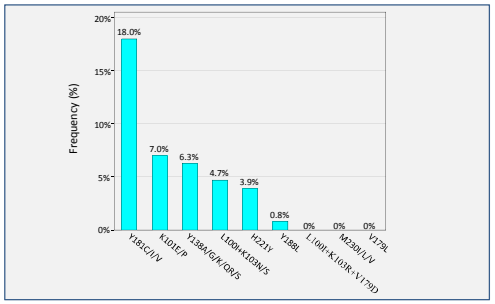

Figure 1: Rilpivirine RAMs

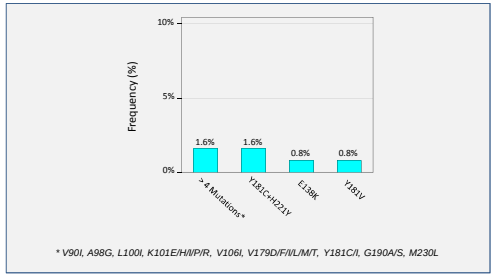

Figure 2: Etravirine RAMs

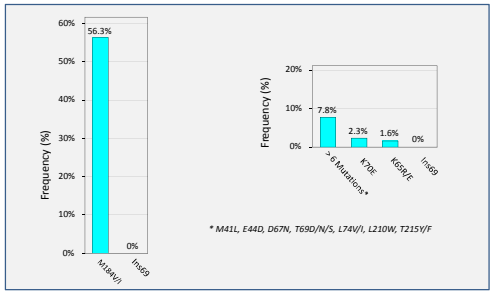

Figure 3: FTC and TDF RAMs

Table 3: FTC/TDF/RPV RAMs

Presence of RAMs (n=128)			
FTC	*TDF*	*RPV*	*N (%)*
Yes	No	No	42 (33%)
Yes	No	***Yes***	21 (16%)
No	No	***Yes***	13 (10%)
Yes	***Yes***	***Yes***	5 (4%)
Yes	***Yes***	No	4 (3%)
No	***Yes***	***Yes***	2 (2%)
No	***Yes***	No	1 (1%)
72 (56%)	**12 (9%)**	**40 (31%)**	**88 (69%)**

Discussion

- RT DNA amplification rate was 76% and independent to baseline CD4 cell count
- Among the patients, all preexposed to NVP/EFV, we only observed 31% and 4% of DNA resistance to RPV and ETR, respectively:
  - Resistance testing performed on HIV DNA lacks sensitivity compared with previous plasma genotypes *(Delaugerre HIV Med 2012, Wirden JAC 2011)*
  - Both RPV and ETR RAMs were most frequently detected in non-B HIV-1 subtype *(Lambert-Niclot JAC 2013)*
- Resistance to FTC/TDF/RPV (Eviplera®) combination was detected among 69% of patients, consistent with M184V alone in 48%

Conclusion

- In patients with suppressed viremia under ART but who had previously failed to efavirenz and/or nevirapine-based regimen, the prevalence of resistance in HIV-1 DNA is 31%, 4% and 69% for rilpivirine, etravirine and the combination of tenofovir/emtricitabine/rilpivirine
- This result confirms that the switch to rilpivirine-based regimen should not be recommended in patients that previously failed to efavirenz and/or nevirapine-containing treatment