

Genotypic Resistance Analysis in Women who received Intrapartum Nevirapine (NVP) Associated to a Short Course of Zidovudine (ZDV) to Prevent Perinatal HIV-1 Transmission: The Ditrame Plus ANRS 1201/02 Study, Abidjan, Côte d'Ivoire

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OBJECTIVE

Ditrame Plus was an open label non randomized trial. Consenting women with HIV-1 infection started oral ZDV (300mg bid) ≥ 36 wks gestation and were treated at the beginning of labor with an oral dose of 600 mg ZDV+200 mg NVP. Neonates were treated with ZDV syrup (2mg/kg/6hrs) for 1 week + a single dose of NVP syrup (2mg/kg on Day 2-3). This substudy evaluates the risk of emergence of NVP resistance mutations at 4 wks postpartum in a sample of women enrolled in this trial and in all infected neonates at wk 4.

POPULATION

402 women were enrolled in the Ditrame Plus trial and 361 were included in the transmission analysis. The overall transmission rate at wk 4 was 6.4% (Dabis F. Barcelona 2002 ThOrD1428). In this substudy, we studied 63 women with samples available at delivery baseline prior to treatment (32 wks of amenorrhoea) and at 4 wk post-partum.

- 21 whose infant was infected (cases)
- 42 whose infant was uninfected (controls). Control maternal specimens were selected in three viral load classes (≥ 3.53 and ≤ 4.2 , ≥ 4.21 and ≤ 4.68 , ≥ 4.69). Samples of the 26 infected children were also studied.

METHODS

Plasma were tested for genotypic resistance analysis, performed by sequencing reverse transcriptase and protease genes. Phylogenetic analysis was performed on the RT gene.

RESULTS

The viral load of the transmitting mothers was significantly higher than the non transmitting mothers (4.89 vs 4.49, $p=0.003$). Twenty-one (33.3%: [21.4%-45.3%]) of 63 women developed a detectable NVP resistance mutation at 4 wk post partum. None of these mutations was present in plasma collected prior to treatment. Frequency of resistant virus was not different in non transmitting mothers (14/42, 33.3%) compared to transmitting mothers (7/21, 33.3%). No ZDV associated mutation was observed.

Six (23%: [8.9%-43.6%]) of the 26 infected children developed nevirapine resistance mutation at week 4.

Id Number	Resistance Mutation at week 4	CD4 /mm ³	Viral load (log copies/ml)
1	103N	85	5.3
2	103N	356	4.21
3	103N	107	4.81
4	103N,179I	328	5.16
5	103N	623	5.01
6	103N	306	5.55
7	103N, 106A, 188C, 190A	307	4.99
8	103N, 181C	333	4.61
9	103N	245	4.93
10	103N, 106A, 188C, 190A	238	4.65
11	103N	336	5
12	103N, 106A	150	5.23
13	103N	111	5.4
14	103N	244	4.95
15	103N	968	3.86
16	106V/A	329	4.66
17	103N	118	5.54
18	103N	270	4.61
19	103N, 106A	71	4.79
20	103N, 181C	614	3.71
21	103N	224	4.2

Table 1: Mutations associated with resistance to nevirapine among the 21 mothers samples at week 4 post partum.

UNIVARIATE ANALYSIS:

Factors associated with the presence of nevirapine resistance mutations in mothers

- Median viral load at inclusion : 4.93 (NVP mutation) vs 4.54 (No NVP mutation), OR [95% CI]: 3.11 [1.04-9.29], $p=0.02$
- CD4 cell count $< 350/\text{mm}^3$: NVP mutation: 81% vs CD4 cell count $\geq 350/\text{mm}^3$: NVP mutation: 19%, $p = 0.06$

Factors not associated with the presence of nevirapine resistance mutations in mothers

- Median viral load at day 2 post partum : 4.09 (NVP mutation) vs 3.91 (No NVP mutation), $p=0.251$
- Median of the difference between viral load at inclusion and viral load day 2 post partum : -0.95 (NVP mutation) vs -1.03 (No NVP mutation), $p=0.980$.

MULTIVARIATE ANALYSIS:

Only the viral load at inclusion was associated with nevirapine resistance mutation, ORa : 3.62 [1.00-13.28], $p=0.05$.

Id Number	Timing of infection	Neonates	Mothers Id	Mothers
A	Intra partum	103N	1	103N
B	In utero	103N/K, 190A/G	2	103N
C	Intra partum	106A	3	103N
D	In utero	103N/K, 106V/A	19	103N/106A
E	In utero	190A/G		No mutation
F	In utero	103N/K		No mutation

Table 2: Mutations associated with resistance to nevirapine among the 6 children samples at week 4 of life. The timing of infection was based on PCR positive at birth (in utero) and PCR negative at birth and positive at week 4 (intra partum)

PHYLOGENETIC ANALYSIS

- Phylogenetic analysis of the 63 HIV-1 strains revealed 78% subtype CRF02, 15% subtype A and 7% subtype CRF06. These results are in accordance with studies previously reported.

CONCLUSION

The incidence of 33% and the patterns of NVP resistance observed in treated mothers predominantly infected with recombinant viruses are comparable to those reported earlier in East Africa. High level of viral replication in mothers is more frequently associated with development of NVP resistance mutations. In those early infected children, treatment with NVP induced acquisition of resistance mutations. The clinical follow-up of the cohort will allow assessment of an eventual persistence of resistance mutation and its impact on response to treatment.