Resistance Patterns Among HIV-Treated Patients With Virological Failure, Busia District, Kenya.

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INTRODUCTION

• **Médécins sans Frontieres Spain (MSF-S) is providing ARV treatment since July 2003 in Busia District Hospital (Kenya).**

• **Busia District, in the western region of Kenya (Western province), has a population of approximately 430,000 and HIV prevalence of 5.9 % (age 15-49 years). After a period of expansion and scale-up of patient numbers, the project is now focusing on strengthening the care and treatment of patients. As of December 2007, approximately 5,000 HIV positive patients have been registered and more than 2,100 of these have been started on ART. The Kenyan MoH aims to reach 75% of patients in need of treatment by 2009.**

OBJECTIVES

• **To assess the usefulness of dried blood spots filter paper (DBS) to determine antiretroviral resistance mutations among HIV+ treated patients with virological failure in Busia District (Kenya).**

• **To ascertain prevalence of genotypic resistance mutations and their pattern.**

• **To determine clinical and epidemiological data among HIV+ patients with virological failure.**

METHODS

Design:

Longitudinal study with immunological and virological follow-up. Genotypic resistance was performed among all patients with virological failure (>400 copies/ml).

Inclusion criteria:

Patients under HAART during ≥12 months (d4T or AZT + 3TC + NVP or EFV). All patients with viral load ≥ 5.000 copies/ml underwent genotypic resistance test.
Non-naïve patients as well as those treated with any other antiretroviral regimen were excluded from the study.

Laboratory:

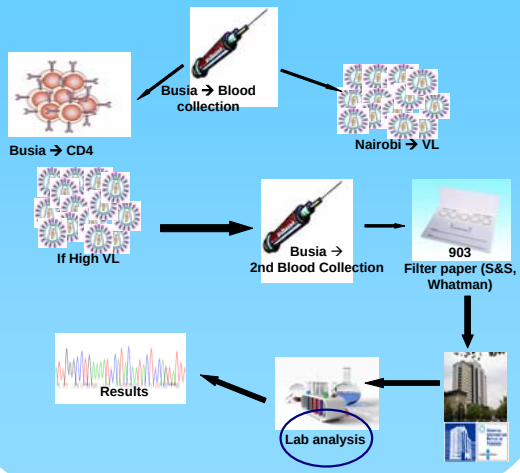
Total HIV-1 RNA from DBS was extracted using Nuclisens method (Easymag, Biomerieux). A fragment of 1023 bp of *pol* gene was amplified using in-house RT-PCR and nested PCR previously described in the literature[1].

Genotyping was validated using the ViroScore Suite (ABL, Luxembourg, v3.9.2).

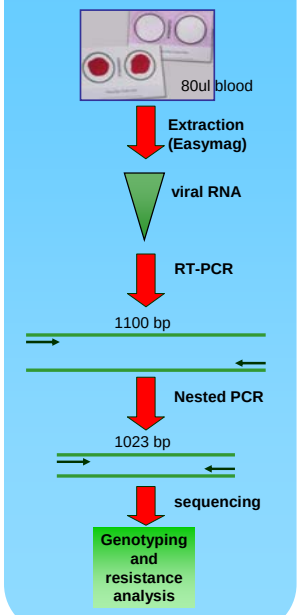
Clinical, epidemiological and adherence data were also collected.

[1] S. Masciotra et al. AIDS 2007, 21:2503–2511

WORKING FLOW



PROCEDURES



RESULTS

•**Eligible patients: 926**
  •**mean treatment duration: 38.8 months**
  •**mean age: 42 y.**
  •**males: 67%**

•**Virological failure: 274 (29.6%)**

•**Viral load > 5000 copies/ml: 55 (5.9%)**

***Table 1.* Patients with VL > 5.000 copies/ml**

VL > 5.000 copies/ml	55/274 (5,9%)
Mean age	37,9 (18,7 – 72,5)
Males	62 %
Clinical failure	18,2%
Median CD4+	306 cells/mm³ (RI: 164 - 469)
Median VL (RNA-VIH)	32.000 copies/ml (IR: 11.000-68.000)

***Table 2.* RAMS among patients with virological failure (VL> 5.000 copies/ml)**

Resistance Associated Mutations (RAMs):	
–Some RAMs	15/18 (83%)
–**TAMS** (M41L, D67N, K70R, V75I, L210W, T215Y)	11/15 (73%)
–**M184V**	15/15 (100%)
–**NNRTI** (K101E, K103N, G190A, V108I, Y181C, Y188L)	12/15 (80%)

RESULTS

ID	HAART	VL	H*	Protease	Retrotranscriptase
2	1	63000	10%	H69K, L89V	K101E, **M184V**, **G190A**, *T215N*
4	1	11000	10%	I62V, L63P, I64V, *H69Y*	**K103N, M184V**
5	1	5600	10%	M36I, D60E	**M41L**, A98G, **K103N, M184V**, **T215Y**
6	1	5700	10%	H69K, *L89M*	**M41L, V75I, V90I, K103N**, *V179I*, **M184V, T215Y**
8	1	1000000	10%	*H69Q*, *L89M*	**M41L**, **V108I**, *V179I*, **Y181C, M184V, L210W, T215Y**
10	1	140000	10%	M36I, I62V, *H69R*, *V82Y*, *L89M*	-----
11	1	9200	10%	D60E, H69K, *L89M*	**M41L, M184V, Y188L**, *L210S*, **T215Y**
13	1	910000	10%	H69K, *L89M*	*V179I*
19	1	6200	60%	*L63Q*, *H69Y*	**K70R, V108I, Y181C, M184V**
30	1	29000	30%	M36I, I62V, *L63T*, I64M, H69K, *L89M*	**M41L, V75I**, *V179I*, **Y181C, M184V, T215Y**
31	1	47000	30%	M36I, H69K, *L89M*	**M41L**, *V179I*, **M184V, G190A, T215F**
33	1	1200000	30%	*E35D*, M36I, H69K, *L89M*	-----
36	1	11000	30%	H69K, *L89M*	*K103S*, *V179I*, **M184V**
37	1	130000	30%	L63P, H69K, *L89M*	**M41L, D67N, K103N, Q151M, M184V, T215F**
38	1	43000	30%	D60E, H69K, *L89M*	**M41L, D67N**, *V179I*, Y181V, **M184V, L210W, T215Y**
39	1	68000	30%	M36I, *L63T*, H69K, V82I, *L89M*, I93L	*K101A*, *V179I*, **M184V, G190A**
46	2	170000	30%	H69K, *L89V*, *L90T*	**M41L, D67N, Y181V, M184V, L210W, T215Y**
53	1	130000	40%	D60E, H69K, *L89M*	**M41L, V108I**, *V179I*, **Y181C, M184V, T215Y**

* % of humidity

***Table 3.* Genotypic results**

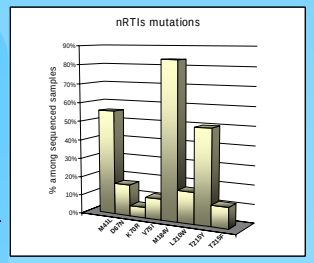


***Figure 1.* NRTI's mutations prevalence**

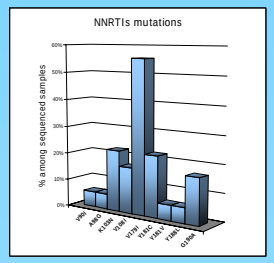


***Figure 2.* NNRTI's mutations prevalence**

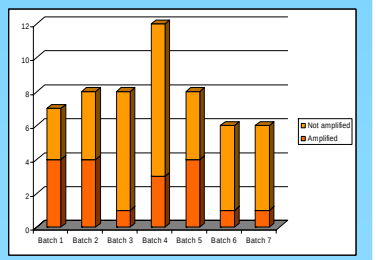


***Figure 3.* Number of amplified samples**

Busia (926 patients; mean Tx duration: 38,8 m)	Malawi[2] (397 patients; mean Tx duration: 9 m)	Cambodia[3] (346 patients; mean Tx duration: 24 m)
VL>400= 16%	VL>400=16%	VL>400= 11.6%
VL > 1000= 10,7%	VL> 1000=13%	VL>1000=9%
VL >30.000= 3,0%	VL>30.000= 5.0%	VL >30.000= 4.3%

***Table 4.* Virological failure rates over time in different resource limited settings**

[2] Ferradini et al. Lancet 2006; 367:1335–42. [3] Ferradini et al. AIDS 2007, 21:2293–2301.

CONCLUSIONS

•**Virological failure rate in resource limited settings are similar to those observed in developed countries.**

•**Overall, genotypic results available were concordant with antiretroviral treatment received by failing patients, reinforcing the importance of treatment adherence in the context of lack of regular access to VL monitoring and genotype.**

•**Our data contribute to the debate whether genotyping are essential and feasible to do in resource poor countries.**

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