Poster Number

605

Scan this QR code to link to this poster and to download a PDF copy.

You will be prompted to enter the following passcode: 605

22nd Conference on Retroviruses and Opportunistic Infections
February 23-26, 2015
Seattle, Washington

Baseline Low-Frequency HIV-1 Variants Do Not Predict Virologic Failure to RPV/FTC/TDF

DP Porter[1*], M Däumer[2], A Thielen[2], MD Miller[1], and KL White[1]

[1]Gilead Sciences, Inc., Foster City, CA, USA
[2]Seq-IT GmbH & Co. KG, Kaiserslautern, Germany

Gilead Sciences, Inc.
333 Lakeside Drive
Foster City, CA 94404
Phone: (650) 574-3000
Fax: (650) 578-9264

Background

- Rilpivirine/emtricitabine/tenofovir disoproxil fumarate (RPV/FTC/TDF) is currently approved for use in HIV-1-infected treatment naïve adults with HIV-1 RNA ≤100,000 c/mL due to increased risk of virologic failure with resistance development in patients with high baseline viral load (>100,000 c/mL)[1,2]
- STaR (GS-US-264-0110) was a phase 3b study evaluating the safety and efficacy of the two single-tablet regimens RPV/FTC/TDF and efavirenz (EFV)/FTC/TDF
  - RPV/FTC/TDF was non-inferior to EFV/FTC/TDF at Weeks 48 and 96[3,4]
  - Through Week 96, 5.3% of subjects on RPV/FTC/TDF and 1.0% of subjects on EFV/FTC/TDF developed resistance by population sequencing[4]
  - Subjects with resistance development by baseline viral load:[4]
    - RPV/FTC/TDF: 9.0% >100,000 c/mL vs. 3.5% ≤100,000 c/mL
    - EFV/FTC/TDF: 0.7% >100,000 c/mL vs. 1.2% ≤100,000 c/mL
- Minority HIV-1 variants present as part of the virus quasispecies at baseline may influence virologic outcome[5,6,7]
- Deep sequencing was performed to assess the potential impact of pre-existing minority variants on virologic outcome and resistance development in:
  - Subjects with resistance development compared to controls
  - Subjects with baseline HIV-1 RNA >500,000 c/mL compared to ≤500,000 c/mL

Methods

Figure 1. STaR Study Design (GS-US-264-0110)

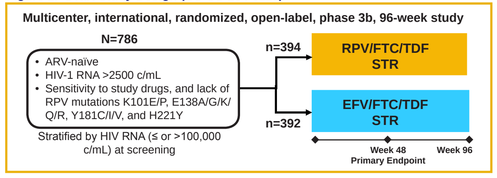
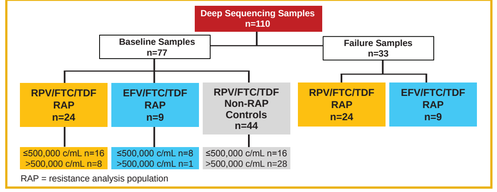


Figure 2. Deep Sequencing Sample Distribution



RAP = resistance analysis population

- 110 samples were analyzed using deep sequencing (Figure 2)
  - 33 baseline and 33 failure samples from subjects that met the criteria for inclusion in the resistance analysis population (RAP; defined below) through Week 96 (24 RPV/FTC/TDF, 9 EFV/FTC/TDF)
  - 44 baseline control samples from the RPV/FTC/TDF arm including samples from all subjects with baseline HIV-1 RNA >500,000 c/mL that did not qualify for the RAP (n=28) and matched controls (baseline HIV-1 RNA and CD4) for subjects in RAP with HIV-1 RNA ≤500,000 c/mL (n=16)
- Criteria for inclusion in the resistance analysis population (RAP):
  - Suboptimal virologic response
    - <1 $\log_{10}$ below baseline at Week 8 and HIV-1 RNA ≥400 c/mL, confirmed at the next scheduled or unscheduled visit
  - Virologic rebound: two consecutive visits with HIV-1 RNA
    - ≥400 c/mL after achieving HIV-1 RNA <50 c/mL OR
    - >1 $\log_{10}$ increase from nadir and HIV-1 RNA ≥400 c/mL
  - Early Discontinuation
    - HIV-1 RNA ≥400 c/mL at last visit at or after Week 8

Methods (cont'd)

- Amplification and deep sequencing (Illumina MiSeq) were performed using the deep*type* HIV assay (Seq-IT)
  - Variants present at frequencies ≥1% are reported
  - Deep sequencing results were compared to population sequencing results (Monogram Biosciences) performed at study entry and virologic failure
  - Pairwise linkage analyses were conducted for subjects with mutation mixtures at RT positions 101, 138, 181, and 184 using only ±150 bp reads containing both positions of interest

Results

Table 1. Baseline Demographics and Characteristics

	RPV/FTC/TDF RAP, n=24	EFV/FTC/TDF RAP, n=9	Non-RAP Controls, n=44
Median age, years (IRQ)	37 (31, 45)	28 (22, 33)	41 (32, 45)
Male	96%	89%	96%
White race	50%	44%	75%
Mean Baseline CD4 cell count, cells/mm³ (SD)	253 (199)	370 (175)	280 (186)
Mean Baseline HIV-1 RNA, $\log_{10}$ c/mL (SD)	5.9 (6.2)	5.4 (5.7)	6.0 (6.2)
≤100,000 c/mL, n (%) >100,000 c/mL, n (%)	11 (46%) 13 (54%)	5 (56%) 4 (44%)	11 (25%) 33 (75%)
≤500,000 c/mL, n (%) >500,000 c/mL, n (%)	16 (67%) 8 (33%)	8 (89%) 1 (11%)	16 (36%) 28 (64%)

Table 2. Summary of NNRTI and NRTI Resistance Mutations Detected in Baseline Samples by Deep Sequencing (≥1%)

	Subjects with mutations present at baseline n (%)			
	Total Baseline n=77	RPV/FTC/TDF RAP, n=24	EFV/FTC/TDF RAP, n=9	Non-RAP Controls, n=44
No mutations	62 (81%)	18 (75%)	8 (89%)	36 (82%)
Any NNRTI-R[a] or NRTI-R[b] ≤500,000 c/mL BL VL >500,000 c/mL BL VL	15 (19%) 8/40 (20%) 7/37 (19%)	6 (25%) 4/16 (25%) 2/8 (25%)	1 (11%) 1/8 (13%) 0/1	8 (18%) 3/16 (19%) 5/28 (18%)
Any NNRTI-R	8 (10%)	3 (13%)	0	5 (11%)
Any NRTI-R	9 (12%)	3 (13%)	1 (11%)	5 (11%)
Both NNRTI-R + NRTI-R	2 (3%)	0	0	2 (5%)

BL VL = baseline viral load
[a] NNRTI resistance mutations (NNRTI-R) are A98G, L100I, K101E/H/P, K103N/S, E138A/G/K/Q/R, V179L, Y181C/I/V, Y188C/H/L, G190A/E/Q/S, H221Y, P225H, F227C, M230I/L in RT.
[b] NRTI resistance mutations (NRTI-R) are M41L, E44D, A62V, K65N/R, D67N, T69D, T69 insertions, K70E/R, L74I/V, V75I, F77L, Y115F, F116Y, Q151M, M184I/V, L210W, T215F/Y, K219E/N/Q/R in RT.

- Baseline NNRTI and NRTI variants were detected in similar proportions of:
  - RAP vs. control subjects
  - Subjects with baseline viral load > vs. ≤ 500,000 c/mL

Table 3. Specific Mutations Detected at ≥1% Frequency at Baseline by Deep Sequencing

Mutation	RPV/FTC/TDF RAP (n=24)		EFV/FTC/TDF RAP (n=9)		Non-RAP Controls (n=44)	
	Number of Subjects (%)	Variant Frequency	Number of Subjects (%)	Variant Frequency	Number of Subjects (%)	Variant Frequency
Any NRTI-R	3 (13%)	Range 1.6->99%	1 (11%)	>99%	5 (11%)	Range 1.1–2.7%
E44D	0	--	1	>99%[b]	0	--
K65R[a]	0	--	0	--	1	2.0%
K70R	1	1.6%	0	--	1	1.5%
V75I	0	--	0	--	2	2.5, 2.7%
F77L	0	--	0	--	1	1.1%
M184V[a]	1	1.0%	0	--	0	--
K219R	1	>99%[b]	0	--	0	--
Any NNRTI-R	3 (13%)	Range 1.0-1.1%	0	--	5 (11%)	Range 1.2-5.3%
E138G[a]	1	1.1%	0	--	1	1.2%
E138K[a]	1	1.1%	0	--	1	2.1%
Y181C[a]	0	--	0	--	1	2.9%
G190E	1	1.1%	0	--	1	3.7%
H221Y[a]	1	1.0%	0	--	0	--
M230I[a]	0	--	0	--	1	5.3%

--, no mutations detected at ≥1% frequency; NNRTI-R = NNRTI resistance mutations; NRTI-R = NRTI resistance mutations
[a] Major RPV/FTC/TDF resistance mutation.
[b] Mutation also detected by population sequencing at baseline; not considered resistant to study drugs.

- All control subjects with baseline low-frequency NNRTI and NRTI resistance mutations were virologic successes at Week 96

Results (cont'd)

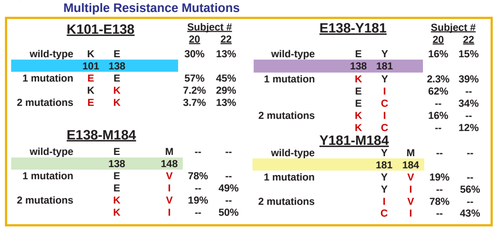
Table 4. Mutations Detected in RPV/FTC/TDF and EFV/FTC/TDF RAP Subjects by Deep Sequencing

Subject Number	Baseline HIV-1 RNA (c/mL)	Failure Type	Baseline		Virologic Failure	
			NNRTI-R	NRTI-R	NNRTI-R	NRTI-R
RPV/FTC/TDF						
1	10500	VR	--	--	E138K (>99)	M184I (>99)
2	12400	VR	--	K219R (99)	E138K (34), E138Q (66), **H221Y (34)**	M184I (>99), K219R (99)
3	20600	VR	--	--	--	**M184V (1.3)**
4	44400	VR	--	--	K101E (43), Y181C (>99), M230L (>99)	M184I (>99), K219E (>90), **V75I (54)**
5	50600	VR[a]	--	--	L100I (>99), K103N (99), P225H (99)	M184V (>99), L74V (>99), **F77L (2.8)**
6	50700	VR	**E138G (1.1)**	--	E138K (>99), H221Y (40)	M184I (>99)
7	59100	VR	--	--	E138K (>99)	M184I (>99)
8	63000	SVR	--	**M184V (1.0)**[d]	Y181C (>99)	**K70E (1.5)**, M184V (>99)[d], **K219E (5.7)**, **K219N (5.6)**
9	65100	VR	--	--	F227C (62)	M184V (63)
10	98900	VR[a]	--	--	--	**D67N (62)**, M184V (35)
11	127000	VR	--	--	E138K (>99)	K70E (>99), M184I (>99)
12	154000	VR	--	--	**Y188C (1.2)**	--
13	215000	VR[b]	--	--	E138K (>99), H221Y (6.1)	M184I (>99)
14	288000	NS[c]	--	--	K101E (99)	Y115F (99), M184I (99), K219E (68)
15	314000	VR	--	**K70R (1.6)**	--	M184I (>99)
16	564000	VR[b]	**H221Y (1.0)**	--	E138Q (>99)	M184I (>99)
17	621000	SVR	--	--	E138K (>99), Y181I (>99)	M184V (>99)
18	891000	VR[b]	--	--	K101E (18), Y181I (99)	M184V (>99)
19	2140000	SVR	--	--	E138K (96), **E138Q (4)**, H221Y (49)	**D67N (2.4)**, M184I (>99)
20	2330000	SVR	--	--	K101E (64), **E138K (11)**, Y181I (85)	M184V (99)
21	2510000	VR[b]	**E138K (1.1)**, **G190E (1.1)**	--	Y181C (99), Y188H (99)	K65R (>99), T69del (24), K219E (>99)
22	6410000	SVR	--	--	A98G (42), K101E (57), E138K (45), Y181C (44), H221Y (42)	M184I (>99)
EFV/FTC/TDF						
1	11200	VR[a]	--	--	Y188L (>99)	**F77L (3.7)**
2	22100	VR	--	E44D (>99)	--	E44D (99)
3	22500	SVR	--	--	--	--
4	32400	VR[b]	--	--	K103N (99)	--
5	97700	VR	--	--	M230L (99)	M184I (>99)
6	150000	VR	--	--	**M230L (>99)**	--
7	161000	SVR	--	--	--	--
8	452000	VR[a]	--	--	--	--
9	1440000	VR[b]	--	--	G190E (55), G190Q (44), **H221Y (8.7)**	**A62V (7.7)**, **K65R (7.7)**, D67N (99), M184I (>99), **K219E (49)**, **K219N (2.4)**

--, no mutations detected at ≥1% frequency; NNRTI-R = NNRTI resistance mutations; NS = never suppressed; NRTI-R = NRTI resistance mutations; SVR = suboptimal virologic response; VR = virologic rebound
Variant frequencies are in parentheses. Mutations not detected by population sequencing are bolded and underlined. Data were not available for failure samples from 2 RPV/FTC/TDF subjects (assay failures).
[a] Virologic rebound was not confirmed. Last visit on study drug.
[b] Subject was never suppressed. Confirmed virologic rebound with >1 log increase in HIV-1 RNA from nadir. Unless noted, other subjects with virologic rebound first achieved HIV-1 RNA <50 copies/mL prior to rebound.
[c] This subject never suppressed but did not meet the criteria for either virologic rebound or non-response; discontinued due to lack of efficacy.
[d] Baseline M184V codon was GTG; virologic failure M184V codon was GTA.

- Low-frequency NNRTI and NRTI resistance mutations detected at baseline were not detected at virologic failure in subjects with resistance development
  - 1 subject in the RPV/FTC/TDF arm with 1.0% M184V at baseline had >99% M184V at virologic failure; however, different codons were present at baseline (GTG) and virologic failure (GTA)
- Deep sequencing at virologic failure detected the presence of additional low-level resistance mutations not detected by population sequencing
  - 2 subjects in the RPV/FTC/TDF arm and 1 subject in the EFV/FTC/TDF arm that did not have resistance mutations by population sequencing at virologic failure had NNRTI or NRTI resistance mutations detected by deep sequencing

Figure 3. Pairwise Linkage Analysis of RT Mutations from Two Subjects with Multiple Resistance Mutations



- Two subjects in the RPV/FTC/TDF arm had mixtures of mutations at RT positions 101, 138, 181, and 181 at virologic failure and were analyzed for pairwise linkage
  - K101E and E138K were most frequently found on different genomes, but did occur together
  - Y181C/I occurred with or without E138K
  - M184I/V occurred alone, or with either E138K or Y181C/I

Conclusions

- **Baseline HIV-1 NNRTI and NRTI resistance mutations were detected in approximately 20% of subjects analyzed by deep sequencing in the STaR study**
  - **Baseline low-frequency variants were not more common in subjects with >500,000 c/mL HIV-1 RNA compared to subjects with baseline HIV-1 RNA ≤500,000 c/mL**
- **Low-frequency HIV-1 variants at baseline were not associated with virologic failure or resistance development in either RPV/FTC/TDF or EFV/FTC/TDF-treated subjects**
  - **Most subjects with low-frequency HIV-1 variants at baseline either achieved virologic suppression or the baseline variants were not detected upon virologic failure**
  - **One subject with 1% M184V at baseline later developed >99% M184V using a different codon, potentially indicating independent resistance development at virologic failure**
- **Deep sequencing results were generally consistent with population sequencing results, but detected additional low-frequency NNRTI and NRTI resistance mutations at virologic failure**
- **Linkage analysis of deep sequencing data showed that M184I/V can occur independently of NNRTI resistance mutations, while NNRTI resistance mutations are observed either separately or in combination with one another**

References

1. Complera US Prescribing Information September 2014
2. Eviplera Summary of Product Characteristics June 2014
3. Cohen et al. AIDS 2014
4. Porter et al. HIV Clin Trials 2015
5. Li JZ et al. JAMA 2011
6. Cozzi-Lepri et al. JAC 2014
7. Van Eygen et al. IHDRW 2012

Acknowledgments

- Ross Martin and Krishna Chodavarapu for analysis of the deep sequencing data
- Patients, study staff, and investigators of the STaR study

© 2015 Gilead Sciences, Inc. All rights reserved.