Poster #0557

HIV-1 RNA Detection By Abbott M2000 Correlates With Integrase Single Copy Assay

Melissa Tosiano[1], Hanna Mar[2], Joshua Cyktor[1], Dianna Koontz[1], Joe Eron[3], Rajesh Gandhi[4], Deborah McMahon[1], Ronald J. Bosch[2], John Mellors[1] for the A5321 Team

[1]University of Pittsburgh, Pittsburgh, PA, USA; [2]Harvard TH Chan School of Public Health, Boston, MA, USA; [3]University of North Carolina, Chapel Hill, NC, USA; [4]Massachusetts General Hospital, Boston, MA, USA

Introduction

- Correlation between labor intensive single copy plasma HIV-1 RNA assays and less sensitive but automated, FDA-cleared plasma HIV-1 RNA assays is not well-defined.
- ACTG A5321 monitors the decay of HIV-1 reservoirs in participants suppressed on long-term antiretroviral therapy (ART).
- The association between a research single copy qRT-PCR HIV-1 RNA assay and the Abbott M2000 commercial platform was investigated by testing entry plasma samples by both methods.
- Correlations between plasma HIV-1 RNA quantified by Abbott M2000 and parameters pre-ART and at study entry were also assessed.

Objectives

- To quantify the correlation of HIV-1 RNA plasma concentration between a research single copy qRT-PCR assay targeting integrase (iSCA) and the automated Abbott M2000 platform.
- To investigate the feasibility of using a high-throughput automated assay in epidemiologic investigations of low-level viremia and to screen for changes in low-level viremia following novel therapeutic interventions.

Methods

- Both assays were performed on 309 plasma samples obtained at a median of 7.3 years after initiation of ART **(Table 1)**.
- Participants were on suppressive ART with plasma HIV-1 RNA below 40 cp/mL by Abbott M2000.
- The single copy qRT-PCR assay targeting integrase (iSCA) was performed as published (Cillo, J Clin Micro 2016).
  - iSCA limit of detection (LoD) 0.4 cp/mL for a 5 mL plasma sample.
  - iSCA results were classified as HIV-1 RNA “detected” or “not detected”.
- The FDA-cleared Abbott M2000 RealTime HIV-1 Viral Load assay was implemented according to manufacturer instructions.
  - Abbott M2000 results were reported as target not detected (<40 TND) or <40 cp/mL target detected but not quantifiable (<40 Target Detected).
- The primary analysis is between the paired results of iSCA (detected or not detected) and Abbott M2000 (<40 TND or <40 Target Detected).
- The secondary analysis is between Abbott M2000 HIV-1 RNA plasma levels and virologic, immunologic and clinical measures pre-ART and at study entry.

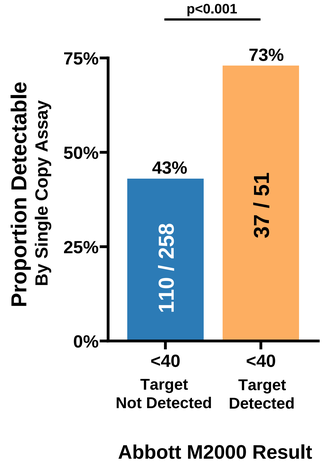
Results

- 52% of iSCA results had undetectable HIV-1 RNA; the undetectable iSCA results were primarily <0.4 cp/mL; nine were <0.5 to <1.1 cp/mL because of lower sample volume.
  - By Abbott M2000, 17% of samples were <40 target detected and 83% were TND.
  - Of the TND by Abbott samples, 43% had HIV-1 RNA detected by iSCA.
  - Of the samples <40 target detected by Abbott, 73% had detectable HIV-1 RNA by iSCA **(Figure 1, Table 3; p<0.001)**.
- Results were similar excluding nine with lower iSCA plasma volume, categorizing iSCA as <0.4 vs. ≥0.4 cp/mL: 44% ≥0.4 cp/mL if TND by Abbott and 73% ≥0.4 cp/mL if target detected (p<0.001).
- There was no evidence of a difference in pre-ART or on-ART measures between participants with HIV-1 RNA <40 TND vs <40 cp/mL target detected by Abbott M2000 at study entry **(Table 2 & 3)** except**:**
  - Cell associated-RNA (CA-RNA) at study entry was modestly correlated **(Table 3).**
  - Women are more likely to be <40 TND than men **(Table 3)**.

Table 1: Baseline Characteristics at Time of Virologic Testing

	Total (N=309)
Age (years)	
Median (IQR)	49 (41, 54)
Sex (N, %)	
Male	253 (82%)
Race/Ethnicity (N, %)	
White, non-Hispanic	172 (56%)
Black, non-Hispanic	61 (20%)
Hispanic (Regardless of Race)	68 (22%)
Asian, Pacific Islander	3 (1%)
American Indian, Alaskan Native	3 (1%)
More than one race	2 (1%)
Years on ART at entry	
Median (IQR)	7.3 (6.1-10.0)
ART regimen at Entry (N, %)	
NNRTI + NRTIs	161 (52%)
PI + NRTIs	84 (27%)
InSTI + NRTIs	63 (20%)
Other	1 (0%)
CD4 cell count (cells/mm^3)	
Median (IQR)	681 (517, 863)

Figure 1: Proportion iSCA Detectable by Abbott M2000



Conclusions

1. **Participants with HIV-1 RNA <40 target detected by a commercial HIV-1 RNA assay (Abbott M2000) had higher proportion of low-level viremia detected by research single-copy assay (iSCA) than those with HIV-1 RNA TND on commercial assay.**
2. **This finding demonstrates that there are differences in low-level viremia between those with target detected and target not detected even when the HIV-1 RNA is <40 on commercial assays.**
3. **Evaluating changes in target detected vs. target not detected using a high-throughput commercial HIV-1 RNA assay during clinical trials may provide early indication of virologic effects of novel therapeutic interventions that should be confirmed with a single-copy research assay.**

Table 2: Effect of Pre-ART Measures on HIV-1 RNA by Abbott M2000

	HIV-1 RNA by Abbott (cp/mL)		
	<40 TND	<40 Target Detected	P-Value
CA HIV-1 DNA (cp/10^6 CD4+ T-cells)			
Median (IQR)	9,685 (4,283-23,035)	12,145 (6,969-53,934)	0.15*
N	80	15	
CA HIV-1 RNA (cp/10^6 CD4+ T-cells)			
Median (IQR)	3,717 (821-32,624)	6,168 (1,192-16,748)	0.90*
N	63	13	
Plasma HIV-1 RNA (log_{10} cp/mL)			
Median (IQR)	4.6 (4.2-5.0)	4.7 (4.5-5.0)	0.36*
N	246	47	
CD4+ T-cell count (cells/mm^3)			
Median (IQR)	260 (111-376)	288 (130-372)	0.63*
N	246	47	

Table 3: Effect of On-ART Measures on HIV-1 RNA by Abbott M2000

	HIV-1 RNA by Abbott (cp/mL)		
	<40 TND	<40 Target Detected	P-Value
iSCA Result (-/+)			
Negative (-) N (%)	**148 (57.4%)**	**14 (27.5%)**	**<0.001**
Positive (+) N (%)	**110 (42.6%)**	**37 (72.5%)**	
CA HIV-1 DNA (cp/10^6 CD4+ T-cells)			
Median (IQR)	516 (193-1,313)	586 (317-1,524)	0.15*
N	237	45	
CA HIV-1 RNA (cp/10^6 CD4+ T-cells)			
Median (IQR)	36 (14-130)	71 (21-165)	0.06*
N	228	45	
IL-6 (pg/mL)			
Median (IQR)	1.5 (1.0-2.1)	1.4 (0.8-2.3)	0.55*
N	246	47	
IP-10 (pg/mL)			
Median (IQR)	118.6 (85.8-159.4)	130.7 (99.2-169.0)	0.20*
N	246	47	
neopterin (nMol/L)			
Median (IQR)	9.1 (7.3-11.1)	9.4 (6.7-11.2)	0.79*
N	246	47	
sCD14 (ng/mL)			
Median (IQR)	1,947 (1,492-2,452)	1,955 (1,251-2,406)	0.60*
N	246	47	
sCD163 (ng/mL)			
Median (IQR)	527 (384-757)	533 (389-803)	0.81*
N	246	47	
TNF-α (pg/mL)			
Median (IQR)	2.0 (1.1-3.3)	1.9 (1.3-3.6)	0.58*
N	246	47	
CD4+ T-cell count (cells/mm^3)			
Median (IQR)	675 (518-877)	700 (507-847)	0.85*
N	246	47	
BMI			
Median (IQR)	26.9 (24.0-31.0)	27.4 (24.3-29.3)	0.99*
N	246	47	
Waist Circumference (cm)			
Median (IQR)	94.5 (87.0-102.4)	92.5 (88-102)	0.90*
N	246	47	
Waist Hip Ratio			
Median (IQR)	0.9 (0.9-1.0)	1.0 (0.9-1.0)	0.27*
N	246	47	
Years on ART at A5321 Entry			
Median (IQR)	7.4 (6.1-10.4)	7.1 (4.7-8.1)	0.14*
N	246	47	
Sex			
Male N (%)	198 (83%)	42 (18%)	0.21**
Female N (%)	48 (91%)	5 (9%)	

* Wilcoxon Rank Sum test ** Fisher’s exact test

The A5321 team wishes to thank the study participants and study staff at ACTG sites. This study was supported by NIH grants UM1AI068636, UM1AI106701, UM1AI068634.